Effects of Starvation on the Frequency of Formation and Width of Growth Increments in Sagittae of Laboratory-Reared Atlantic Menhaden *Brevoortia tyrannus* Larvae

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Abstract.– We investigated increment formation in sagittae of Atlantic menhaden *Brevoortia tyrannus* in laboratory experiments, and found that the age of individual larvae can be estimated within ±3 days over the first month of life using counts of growth increments. Sagittae were first observed during embryonic development. The first prominent growth increment was formed at first feeding, and the frequency of increment formation of fed and starved larvae ranged from 0.86 to 0.98 increments per day thereafter. Starvation did not appear to systematically alter the periodicity of increment formation from one increment per day, although it consistently modified the width of growth increments among different age groups of larvae. Microstructural growth patterns in sagittae responded rapidly (days) to changes in feeding: larvae starved for 1–3 days formed narrow, poorly-defined increments compared with fed larvae that formed wide, well-defined increments. Standard length and estimated dry weight of larvae were related to sagittal radius by asymptotic and logistic functions, respectively. Sagittal radius of larvae was related to days after first feeding by a logistic function. Our results for Atlantic menhaden confirm the potential of otoliths in providing information about age, stressful events, and growth history of individual fish larvae.

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Rates of growth and survival of young fish are hypothesized to affect the abundance of the incoming year-class (Lasker 1985, Rothschild 1986). Both biotic (e.g., prey resources) and abiotic (e.g., water temperature) factors have a direct effect on the growth and survival of freshwater and marine fish larvae. Microstructural growth patterns in otoliths of teleost fish may provide a record of environmental and physiological condition throughout the larval and juvenile stages and hence important information about processes regulating recruitment in fish (Pannella 1980, Houde 1987, Rice et al. 1987).

The examination of microstructural growth patterns in otoliths for making inferences about the ecology of young fishes has become a popular technique since Pannella (1971) postulated that annuli (yearly growth zones) consisted of growth increments formed on a daily basis. Subsequently, microstructural growth patterns have been used to estimate age and growth histories of fish (Methot and Kramer 1979, Penney and Evans 1985), infer the temperature chronology of larval and juvenile life stages (Radtke 1984, Gauldie et al. 1986), detect life history transitions (Brothers and McFarland 1981, Campana 1984a), and investigate patterns of recruitment and mortality (Crecco et al. 1983, Essig and Cole 1986) and stock identification (Mulligan et al. 1987).

Age validation studies of larval and juvenile fishes have shown that microstructural characteristics are species-specific and may be influenced by nutrition and/or environmental variables (Campana and Neilson 1985, Rice et al. 1985, Jones 1986). Fish larvae subjected to periods of stress (e.g., starvation) or cyclic environmental variables (e.g., diel fluctuations in water temperature) may have their increment deposition disrupted, resulting in apparent nondaily formation (Taubert and Coble 1977, Jones 1984, Neilson and Geen 1985). These results suggest that validation studies are necessary for a species before analysis of otolith microstructure can be used to age individuals in nature. Substantial errors may be incorporated into the analysis if daily increment formation is assumed but nondaily deposition occurs (Campana and Neilson 1985).

Few studies have assessed the effect of short-term starvation on the accuracy of age estimates and growth histories of the early life stages of fish derived from the analysis of otolith microstructure. This paper examines the reliability of sagittal microstructure of larval Atlantic menhaden to estimate age, detect stressful events, and infer the growth chronology of individual larvae. Two age groups of laboratory-reared larvae were subjected to short periods of starvation and optimal feeding conditions to determine the relationship between age, growth history, and microstructural growth patterns in sagittae.

The Atlantic menhaden is a commercially and ecologically important species along the Atlantic east coast (Reintjes 1969, Reish et al. 1985). Recruitment of this species has undergone marked fluctuations since monitoring of the fishery began in the 1940's (Ahrenholz et al. 1987). Investigations of the early life stages of Atlantic menhaden indicate that physical and biological factors operating during larval drift may influence the growth and survival and hence recruitment (Nelson et al. 1977, Checkley et al. 1988). Information derived from these laboratory experiments will serve as a basis for interpreting the microstructure of sagittal otoliths of Atlantic menhaden larvae collected in nature (Maillet 1988).

Methods

Spawning and rearing conditions

Fertilized eggs (henceforth called the "stock population") were obtained from an induced spawn of a captive stock (Hettler 1981, 1983) and placed into three circular tanks containing 60-L of filtered (20 μm) seawater. Eggs and larvae were incubated at 19°C (18.9 ± 0.1°C; $\bar{x} \pm$ SE) in lightly aerated, static water with overhead fluorescent lighting on a 12 L:12 D photoperiod. Salinity ranged from 29 to 33 g/kg. Furan II (7 mg/L, Aquarium Pharmaceuticals) was added to retard the growth of bacteria and fungi. During early development (first feeding to 12 days postfertilization), larvae were offered cultured algae *Nanochloris* spp., rotifers *Brachionus plicatilis*, and wild microzooplankton (70–250 μm size range) ad libitum. During later development, larvae were offered only wild microzooplankton. Dead larvae and settled plankton were removed every 1–3 days. Water level was maintained by removal of seawater each time food was added.

Reference to trade names does not imply endorsement by the National Marine Fisheries Service, NOAA.

Experimental procedures

Eggs were sampled daily during development and preserved in 95% ethanol to investigate otolith formation in embryos. A total of 15 eggs were used in the analyses. To estimate the time to first increment formation and to test for subsequent daily increment formation, 10 to 15 larvae were sampled from the stock population at 3, 4, 5, 6, 7, 8, 11, 14, 25, 34, and 35 days postfertilization. The effect of starvation on the periodicity of increment formation was investigated by exposing two different age groups of larvae (13–20 days, and 28–36 days postfertilization) to short periods with no food. For each group, larvae of various sizes were randomly selected from the stock population and transferred to eight 10-L experimental tanks filled with 20-μm filtered seawater. Initial densities of larvae in experimental tanks were 5 larvae/L (13–20 days) and 4 larvae/L (28–36 days). Environmental conditions, including illumination, water temperature, salinity, and the use of Furan II, were identical to those of the stock tanks. Four experimental groups, consisting of (a) continuous feeding (controls), (b) 1-day starved, (c) 2-day starved, and (d) 3-day starved treatments, were randomly assigned to duplicate experimental tanks. Control larvae were fed wild microzooplankton ad libitum immediately after transfer; treatment larvae received food at the end of the respective starvation interval and were allowed to continue feeding for several days after this period.

All larvae sampled from the stock and experimental containers were first anesthetized with tricaine methanesulfonate (Cresent Research Chemical), measured for standard length (tip of upper jaw to end of notochord) to the nearest 0.1 mm, and then preserved in 95 % ethanol. Larvae were stored individually in 10-mL vials. The preservative was changed once after 48 hours to maintain sagittae in optimal condition.

Otolith preparation and analysis

Sagittae were examined within two months after preservation. The right sagitta was teased from the inner ear with minuten needles, cleaned of excess tissue, and mounted medial-side-up in Flo-Texx (Lerner Laboratories). Specimens were examined with transmitted light under a compound microscope fitted with a 100× objective and a video camera and monitor, thereby increasing the total magnification (monitor image/actual size) to 3600×. An electronic caliper was used to measure growth increments on the video monitor. This system allowed electronic enhancement of otolith images (e.g., contrast between the incremental and discontinuous zones).

Increment counts were made in triplicate on masked (i.e., of unknown origin) samples, to minimize bias, and

the mean calculated. If triplicate counts deviated by ±3 increments or more, the specimen was excluded from further analysis. Five percent of the samples were thereby excluded. Another 16% of the specimens were excluded from the analysis because of our inability to resolve increments in some specimens and/or improper orientation for viewing specimens. Linear regressions of mean increment count on days after first feeding were computed for each experimental group. Student's t was used to determine whether increment formation is initiated at first feeding (i.e., regression intercept = 0) and growth increments are formed daily (i.e., slope = 1.0). Statistical power to detect a deviation of 0.1 from a slope of 1.0 at $p = 0.05$ level (two-sided test) was estimated for each linear regression (Rice 1987, Steel and Torrie 1980). Analysis of variance was used to test the homogeneity among all slopes.

The width of growth increments formed before, during, and after starvation was measured for 5–10 larvae from each experimental tank. Increment width was measured from the perimeter moving inward along the maximal radius, the inner end of the radius being defined as the center of the nucleus. This line was consistently the best for enumerating increments. To eliminate bias in measurement of increment width the focal plane was adjusted, if necessary, for the measurement of each increment. Increment width was averaged for each larva over each of the three treatment intervals (i.e., fed, starved, refed), except for the 1-day starved treatment during starvation. Analysis of covariance (ANOCOVA) was used to compare the mean width of growth increments between larvae from the control and treatment tanks during and after starvation (Steel and Torrie 1980). Since the duration of the starvation and recovery intervals varied with length of starvation, the ANOCOVA tested for differences in mean increment width between larvae from control and treatment tanks within intervals. The interval duration was identical between comparisons of mean increment width of larvae from the control and treatment tanks. Comparison of increment width between fed and starved larvae within intervals was necessary because of age-related trends in increment width. The covariate included in the ANOCOVA was mean increment width prior to the starvation and recovery intervals. The ANOCOVA took into account any differences in the width of growth increments of larvae allocated to the experimental tanks and provided a more accurate comparison between individuals from control and treatment groups.

The relationships between sagittal radius, standard length, estimated dry weight, and days after first feeding were also investigated. We pooled all of the larvae in the treatment groups (i.e., 1-, 2-, and 3-day starved larvae) for analysis, except that starved larvae were excluded when estimating the dry weight—otolith size relation since these measurements were not made directly on individual larvae. Sagittal radius was measured from the center of the nucleus to the perimeter along the maximal radius. Dry weight of individual larvae was estimated from a linear regression of log-transformed dry weight (DW, μg) on log-transformed standard length (SL, mm): $\ln DW = -3.041 + 3.799 \ln SL$, $n = 195$, $r^2 = 0.95$. This equation was derived for laboratory-reared Atlantic menhaden larvae ranging in size from 5 to 25 mm (Checkley et al., ms in prep.). Linear and nonlinear regressions of standard length and estimated dry weight on sagittal radius, and sagittal radius on days after first feeding were compared to determine the best predictive models. Average growth rates (AVG, mm/day) of larvae for both age groups were estimated from first feeding (ff) to experiment termination by: $AVG = (SL - 4.7)/(\text{days after ff})$, where 4.7 is the average standard length (mm) at first feeding (Powell and Phonlor 1986).

Results

Increment description and larva growth rate

Sagittae were first observed during embryonic development and consisted of the dark and apparently proteinaceous primordium (Fig. 1). Examination of sagittae with the light microscope revealed that no growth increments were formed during this period. At hatching, the sagitta resembled a flattened spheroid or hemisphere with a mean radius of 4.8 ± 0.3 μm, $n = 15$. The first increment surrounding the primordium was typically characterized by a wide discontinuous zone and coincided with hatching. In some cases, 3–4 narrow, poorly defined increments were observed outside the first increment. Limited resolution of the light microscope did not always allow these increments to be resolved, counted, and/or measured. The first prominent increment in sagittae formed 3–4 days after hatching and coincided with the initiation of exogenous feeding. This prominent growth increment was used as the starting point for subsequent counts along the maximal radius.

Mean size at hatching for this laboratory population of Atlantic menhaden was 3.59 ± 0.22 mm SL, $n = 100$ (range 3.08–4.00 mm). The average growth rate of menhaden larvae from first feeding to experiment termination was 0.368 ± 0.006 mm/day (13–20 days) and 0.360 ± 0.004 mm/day (28–36 days). Size range for the two age groups examined was 6.4–13.0 mm and 10.3–20.0 mm SL. This rate of growth slightly exceeded the estimate of 0.32 mm/day based on data of Powell and Phonlor (1986) for Atlantic menhaden larvae reared at 20°C. Larvae did not metamorphose

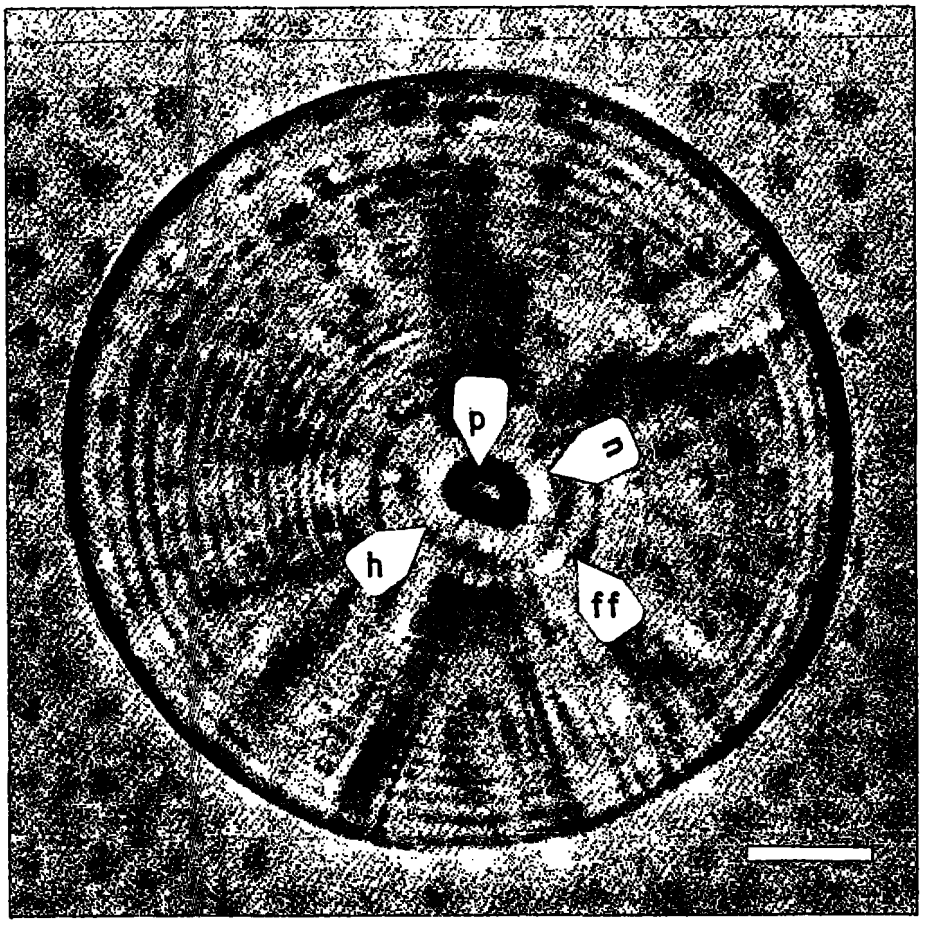

Figure 1
Light micrograph of sagittal otolith of a 22 day-old (postfertilization) laboratory-reared Atlantic menhaden larva showing 18 growth increments. The primordium (p) is delineated by an innermost protein-rich layer. The nucleus (n) is delineated by the first continuous growth increment surrounding the primordium. The first growth increment formed at hatching (h) is characterized by a thick protein-rich layer, and the first prominent growth increment is formed at first feeding (ff). Scale bar represents 10 μm.

in this experiment, but estimates in the literature indicate that wild Atlantic menhaden transform to the juvenile life stage at 25–30 mm (Lewis et al. 1972, Nelson et al. 1977).

Initiation and frequency of increment formation

The time to first increment formation and frequency of increment deposition were estimated from linear regressions of mean increment count on days after first feeding. The regression intercepts of larvae from the stock population and control and treatment groups were not significantly different from zero, except for the 1-day starved treatment, indicating that the first prominent increment is formed at or near the time of first feeding (Table 1).

Frequency of increment formation varied from 0.86 to 0.98 increments/day among experimental groups (Table 1, Fig. 2). There did not appear to be any systematic effect of starvation on frequency of increment formation. The rate of increment formation for both the control and 1-day starved treatments were significantly different from one increment/day, but the stock population, 2-, and 3-day starved treatments formed increments at a rate not significantly different from one increment per day. Computations of statistical power indicated low variability among the increment count—age regressions and provided additional confidence that growth increments are formed daily. The test for homogeneity of slopes indicated a significant difference between larvae from the control and pooled treatments ($p<0.05$, $n=108$). The difference between the estimated slopes for individuals from the control and 1-day starved treatment tanks accounted for this difference. The estimated pooled slope for the control and 2- and 3-day starved treatments was not significantly different in the test for homogeneity among slopes ($p>0.63$, $n=68$).

Response of increment width to changes in larva feeding

Since there were no significant differences in increment width between duplicate containers within different levels of treatments ($p>0.05$, ANOVA), measurements

Table 1

Least-squares regressions of mean increment count on days after first feeding (*ff*) of laboratory-reared Atlantic menhaden: Increment count = $a + b$ (days after *ff*). Asterisks denote significant deviations from the hypothesized values (H_0: $a = 0$, $b = 1.0$, Student's t: $^*p<0.05$, $^{**}p<0.01$, $^{***}p<0.001$).

Experimental group	n	Intercept $a \pm$SE	Slope $b \pm$SE	r^2	95% CL[1]	Power[2]
Stock	79	-0.01 ± 0.11	0.98 ± 0.01	0.99	± 1.8	0.99
Control			*			
0-day starved	27	0.73 ± 0.63	0.92 ± 0.03	0.97	± 2.4	0.90
Treatments		***	***			
1-day starved	40	2.15 ± 0.39	0.86 ± 0.02	0.98	± 2.2	0.99
2-day starved	20	-0.19 ± 0.58	0.96 ± 0.03	0.98	± 2.4	0.91
3-day starved	22	-0.47 ± 0.74	0.94 ± 0.04	0.97	± 2.6	0.82

[1]95% confidence limits (CL) for estimating increment count of an individual larva from time of first feeding.
[2]Estimate of statistical power to detect a deviation of 0.1 from a slope of 1.0 at the $p = 0.05$ level.

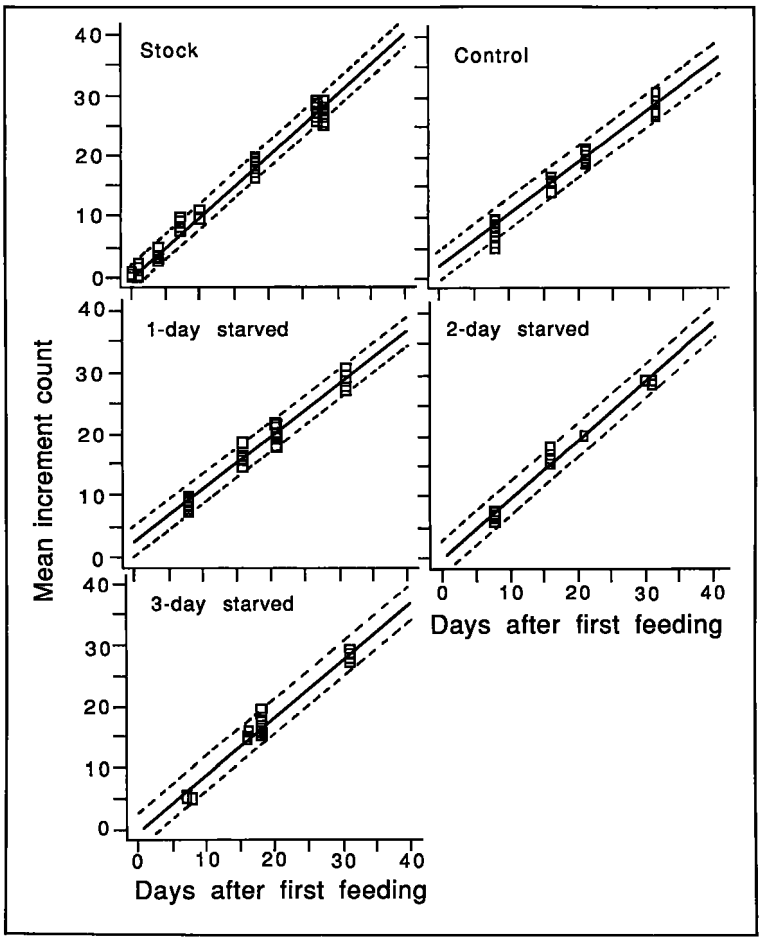

Figure 2

Least-squares regression (—) of increment count on days after first feeding and 95% confidence limits (----) for estimating mean increment count of an individual Atlantic menhaden larva. Some symbols represent more than one observation. Statistics are given in Table 1.

of increment width of larvae from duplicate control and treatment tanks were pooled. Prior to starvation, no significant differences ($p>0.05$) were observed in mean increment width between larvae from control and treatments except for the control and 3-day starved treatment in the older age class. The mean width of growth increments differed significantly ($p<0.05$) between fish from control and treatments during the respective periods of starvation and recovery (Fig. 3). The width of growth increments was significantly larger in fed compared with starved larvae. In general, the magnitude and significance of these differences

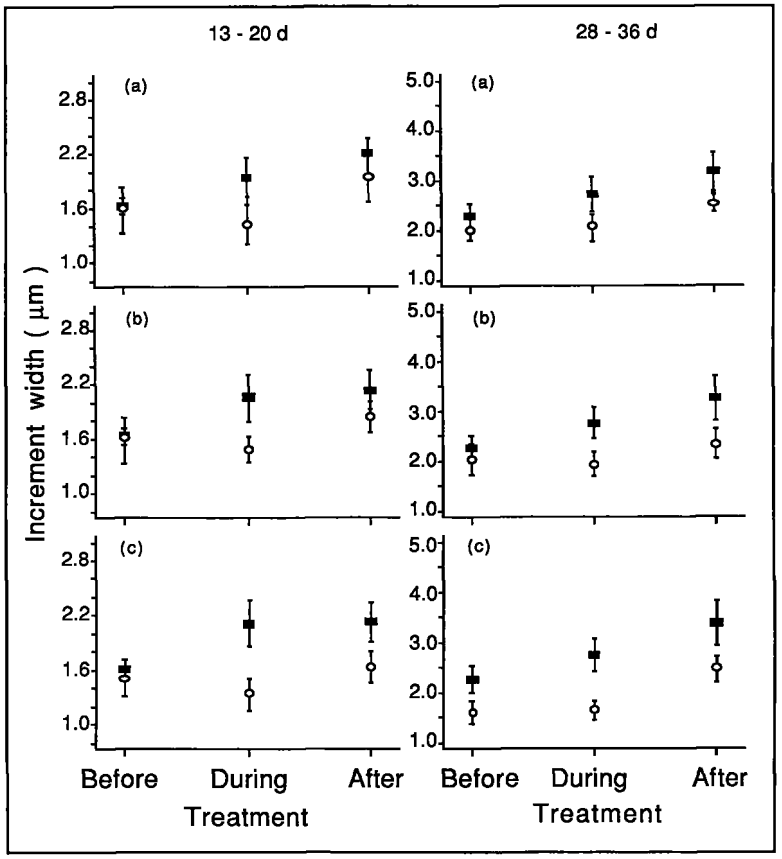

Figure 3
Mean increment width of Atlantic menhaden larvae from control (fed, ■) and treatment (1–3 day starved, O) tanks before, during, and after the respective starvation interval. (a) 1-day starved, (b) 2-day starved, and (c) 3-day starved larvae. Vertical bars represent standard errors.

increased with longer starvation, and both age classes of larvae responded similiarly to these stressful events. Starved larvae in both age classes displayed narrow [1.4 ± 0.1 μm, $n = 60$, (0.7–2.2 μm, range), 13–20 days; 1.8 ± 0.3 μm, $n = 63$, (1.0–3.2 μm), 28–36 days] poorly defined increments while control larvae exhibited wider [2.1 ± 0.2 μm, $n = 30$, (1.2–2.8 μm), 13–20 days; 2.7 ± 0.6 μm, $n = 57$, (1.3–4.6 μm), 28–36 days] well-defined increments during the starvation interval. After starvation, mean increment width of starved larvae in both age classes increased during the 3–6 day recovery interval (Fig. 3).

The results of the ANOCOVA, which adjusted for mean increment width prior to the initiation of starvation, indicated that mean increment width differed significantly ($p < 0.05$) between individuals from control and treatment tanks during starvation in both age classes (Table 2). This result suggests that differences in mean increment width arose during the starvation interval and were not due to differences prior to this interval. However, during the recovery interval, no significant differences ($p > 0.05$) were observed between larvae from control and treatment tanks when adjusted by the covariate. This result suggests that differences observed between individuals from control and treatments during the recovery interval were the result of differences formed during the starvation interval, and indicates that increment width of larval Atlantic menhaden responds rapidly to short-term variations in feeding.

Relationship between sagittal size and body size and sagittal size and age

Visual inspection of residuals for regressions of standard length and estimated dry weight on sagittal radius, and sagittal radius on days after first feeding, indicated that nonlinear models were superior to linear models in all cases. Asymptotic regressions of standard length on sagittal radius were fit separately for larvae from control and pooled treatments (Fig. 4a). The regressions were highly significant ($p < 0.0001$) and residuals were distributed at random over the entire size range examined, indicating that the models fit the data well. The regression coefficients for fed larvae were slightly larger than for starved larvae. A logistic function was used to regress estimated dry weight on sagittal radius (Fig. 4b). Data were log-transformed to stabilize the variance and reduce the influence of larger values on the regression. Starved larvae were excluded from this relationship since dry weights were estimated from standard length. The regression was highly

Table 2

Comparison of least-squares means (adjusted for the covariate in the ANOCOVA) of increment width during and after starvation between control (fed) and treatment (starved) Atlantic menhaden larvae aged (a) 13–20 days, and (b) 28–36 days. N refers to the number of larvae examined, while n corresponds to the total number of observations of increment width from which the mean was calculated. Asterisks denote significant deviations between larvae from control and treatment tanks; * $p<0.05$, ** $p<0.01$, *** $p<0.001$.

			Mean increment width (μm)	
Experimental group	N		During treatment $\bar{x} \pm$ SE (n)	After treatment $\bar{x} \pm$ SE (n)
(a)				
Control	10	*	1.89±0.13 (10)	2.08±0.12 (69)
1-day starved	10		1.48±0.13 (10)	2.01±0.12 (70)
Control	10	**	2.05±0.11 (20)	2.00±0.10 (59)
2-day starved	10		1.49±0.11 (20)	1.89±0.10 (60)
Control	10	***	2.12±0.12 (30)	1.94±0.11 (49)
3-day starved	10		1.33±0.12 (30)	1.81±0.11 (50)
(b)				
Control	19	*	2.63±0.09 (19)	2.97±0.12 (109)
1-day starved	19		2.28±0.12 (13)	2.73±0.14 (84)
Control	19	***	2.68±0.10 (38)	2.73±0.14 (90)
2-day starved	18		2.03±0.14 (12)	2.97±0.19 (42)
Control	19	**	2.53±0.10 (57)	2.94±0.10 (71)
3-day starved	10		2.03±0.14 (30)	3.32±0.16 (38)

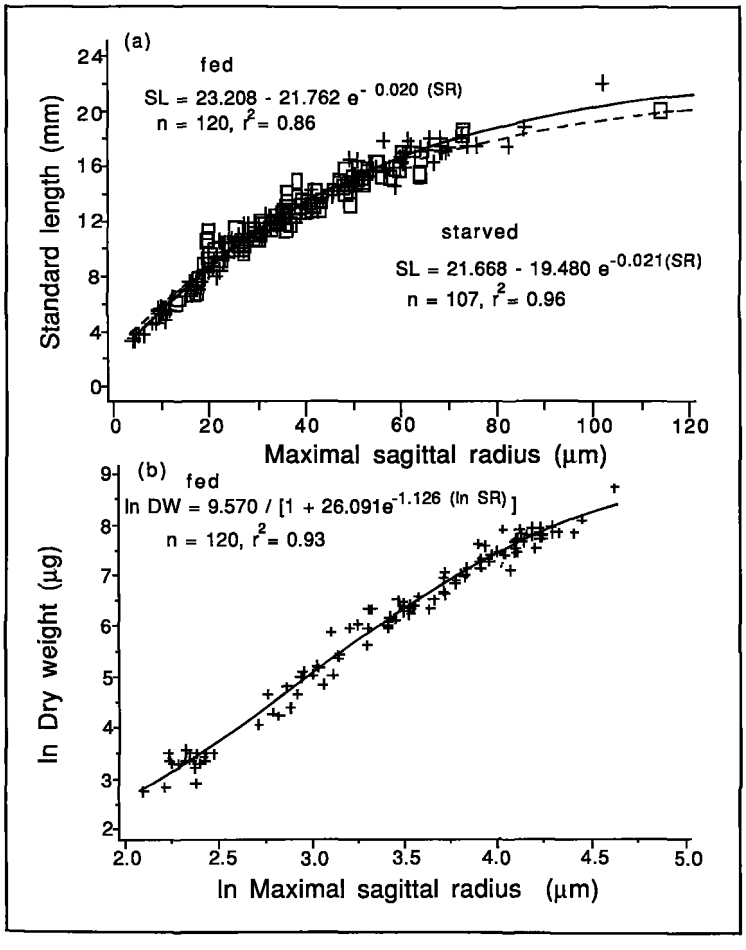

Figure 4

(a) Nonlinear regressions of standard length (SL) on sagittal radius (SR) of Atlantic menhaden larvae from control (solid line, +) and treatment (1–3 day starved, broken line, □) tanks. (b) Nonlinear regression of log-transformed estimated dry weight (DW) on log-transformed sagittal radius (SR) for control (+) larvae only. Model coefficients and other descriptive statistics are also presented. Some symbols represent more than one observation.

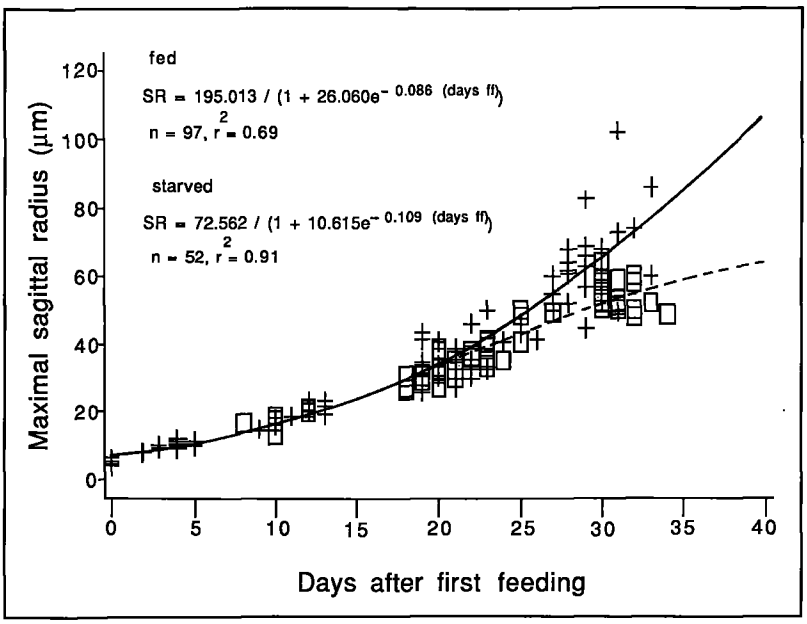

Figure 5
Nonlinear regression of sagittal radius (SR) on days after first feeding (days *ff*) of Atlantic menhaden larvae from control (solid line, +) and treatment (1–3 day starved, broken line, □) tanks. Model coefficients and other descriptive statistics are also given. Some symbols represent more than one observation.

significant ($p<0.0001$) and residuals were distributed at random over the entire size range examined, indicating a good fit of the logistic model to the log-transformed data. The relationship between sagittal radius and days after first feeding was also fit with a logistic function (Fig. 5). The regressions were highly significant (<0.001) and residuals were distributed at random, but the variance increased with age of the larvae. Predictions of sagittal radius from the regressions indicated that increment width increased from 0.6 µm at first feeding to 3.8 µm at 30 days in larvae from the control tanks, while increment width increased from 0.7 to1.6 µm in larvae from the treatment tanks.

Discussion

The results of our laboratory experiments indicate that microstructural growth patterns in sagittae of Atlantic menhaden can be used to accurately estimate age from first feeding, detect short-term variations in growth rate caused by starvation, and estimate the growth chronology of individual larvae. Age of individual larvae from first feeding can be estimated within ±3 days during the first month of life (based on inverse regression: Days after first feeding = a + b (increment count); Draper and Smith 1966, Rice 1987). Atlantic menhaden larvae appear to initiate increment formation at hatching, although growth increments formed prior to first feeding are narrow (<1 µm), poorly defined, and could not be consistently resolved with light microscopy. Poorly-defined increments observed during yolk feeding in the Atlantic menhaden have been observed during similiar periods in other species (Lough et al. 1982, McGurk 1984, Bolz and Lough 1983). The formation of these increments may be related to an immature circadian rhythm (Campana 1984b) and/or the presence of increments too narrow to detect using light microscopy (Campana et al. 1987). Formation of the first prominent growth increment in sagittae at first feeding may be related to the shift to exogenous feeding and related circadian activity patterns.

Transitions within the egg and larval stages of Atlantic menhaden were characterized by particular microstructural features. Hatching was characterized by a wide discontinuous zone, and the transition to exogenous feeding coincided with a prominent growth increment. Similiar patterns have been observed in other species (Brothers and McFarland 1981, Campana 1983, Lagardere and Chaumillon 1988). Particular microstructural features formed during the early life stages may be the result of changes in physiological metabolism, stress, and/or growth cycles that are generally associated with these transitions.

Studies that have examined the rate of increment formation in fish larvae reared under conditions promoting rapid growth have shown that growth increments form daily in most cases (Jones 1986). Atlantic menhaden larvae fed ad libitum formed increments at a rate of unity, consistent with the hypothesis that increments are formed daily in sagittae of well-nourished fish larvae. Estimates of statistical power obtained in our study indicate low variability about the relationship between increment count and days after first feeding and provide additional support that growth

increments are formed daily in sagittae of Atlantic menhaden larvae. A closely related species, the Gulf menhaden *B. patronus*, initiated increment deposition at first feeding and formed an average of one growth increment per day in sagittae (Warlen 1988). The frequency of increment formation in juvenile Atlantic menhaden held in enclosures also indicated that sagittal growth increments are formed daily (Simoneaux and Warlen 1987).

Starvation of larval Atlantic menhaden for 1–3 days did not result in the cessation of sagittal growth nor systematically alter the periodicity of increment formation from one increment per day. Larvae starved for 2- and 3-days formed increments at rates similiar to larvae which were fed continuously. Similiar results have been reported for other species reared under various feeding regimes, including starvation (Marshall and Parker 1982, Campana 1983, Eckmann and Rey 1987). However, other studies investigating formation of otoliths have found that periods of starvation may affect the rate of increment formation (Townsend and Graham 1981, Geffen 1982, McGurk 1984, 1987). Growth increments formed during starvation and periods of slow growth may be difficult to resolve due to their small size , generally <1 μm, which is near the limit of resolution of most light microscopes. This may have resulted in underestimation of the number of increments in two of the experimental groups of Atlantic menhaden larvae in our study and contributed to the apparent nondaily deposition rate. Theoretical daily increment width of larval Atlantic herring during early growth (first 2 weeks) was predicted to be below the limit of resolution of light microscopy (Campana et al. 1987). Examination of otoliths of fish exposed to suboptimal conditions indicated that age was underestimated using light microscopy compared with high resolution viewing by SEM (Jones and Brothers 1987, Bailey and Stehr 1988).

Microstructural growth patterns observed in sagittae of larval Atlantic menhaden support the hypothesis that variations in nutrition and environmental factors affecting fish growth are manifest as variations in increment width (Methot and Kramer 1979, Neilson and Geen 1982, 1985, Radtke 1987). The response of increment width to short intervals of starvation indicate that sagittae of Atlantic menhaden larvae may provide a reliable record of short-term (e.g., days) changes in feeding. The response time to starvation of Atlantic menhaden larvae examined in our study was rapid. Increment width of larvae starved for only 1 day declined significantly compared with fed larvae. The response time of increment width to cessation of starvation varied with the age of larvae. Mean increment width of larvae in the young age class increased to that of the controls within 3–4 days after starvation. Mean increment width of larvae in the older age class increased less rapidly after starvation than did that of the younger age class and did not reach that of controls during the recovery interval (4–6 days).

Our results indicate that microstructural growth increments in sagittae of Atlantic menhaden can be used to infer the dynamics of larval growth. In particular, examination of sagittal microstructure can be used to estimate the age of individual larvae from first feeding, detect stressful periods, and reconstruct the growth rate chronology of Atlantic menhaden larvae. We are currently investigating short-term (e.g., days) variations in growth rate of sea-caught Atlantic menhaden larvae, inferred from analysis of sagittal microstructure, in relation to meteorological and oceanographic variables (Checkley et al. 1988, Maillet 1988). Measurements of individual growth increments, although tedious to obtain, may allow inference about important events during the early life stages of fish. Variations in the width of growth increments from otoliths of fish larvae collected in nature could be analyzed for correlations with the timing of particular developmental and environmental events to determine their relative importance.

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