Abstract. – Studies were performed to determine effects of environment and physiology on the formation of daily increments in winter flounder otoliths. Otoliths from embryonic to 1-yr-old laboratory-raised winter flounder *Pleuronectes americanus* and young-of-year wild-caught specimens were examined, and growth patterns were determined from photographs taken on light and scanning electron microscopes. Behavioral observations were made from hatching through metamorphosis.

Daily growth increments of otoliths from larval winter flounder were enumerated, and a growth curve was derived describing the first 2 months of life. Growth was best described by a Gompertz-type curve. The relationship between sagitta size and fish length was exponential for larvae, but linear during the remainder of the first year. Sagittae were compared with fish length for both wild and laboratory-reared juveniles and exhibited the same relationship for each. The change in relationship between sagitta size and fish length coincided with changes in dimensional growth of the fish.

During metamorphosis, swimming and feeding modes changed from tail-propelled, upright swimming and frequent sudden feeding lunges in larvae, to bottom-resting and creeping accompanied by infrequent feeding gulps in juveniles. This change reflected the transfer from pelagic to benthic habitat and anatomical transformation to asymmetrical form. In general, juveniles maintained lower activity levels than did larvae. Behavioral and anatomical changes are summarized.

Early growth, behavior, and otolith development of the winter flounder *Pleuronectes americanus*

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Many aspects of fish development are reflected in otolith structure. Short- and long-term changes in growth rate may be caused by either environmental fluctuations or life history changes (e.g., metamorphosis, spawning), and these events may also be incorporated into the otolith record of sagittae. Hyaline bands have been used for decades to estimate age. Daily growth increments have also been discovered in fish otoliths (Pannella 1971, 1974) and are proving a powerful tool to study larval population dynamics.

One daily growth increment includes both a calcium-rich aragonite layer in a protein-poor matrix (the "incremental zone") and a poorly calcified protein-rich matrix layer (the "discontinuous zone") (Watabe et al. 1982). Increments are entrained in response to a 24 h light/dark cycle (Taubert & Coble 1977, Tanaka et al. 1981, Radtke & Dean 1982) as well as influenced by other cues (Campana & Neilson 1982, Campana 1984a,b). As a result of the daily cycle in calcium deposition, otoliths often reflect fish age, irrespective of growth, although this has not always been found to be the case (Geffen 1982, Campana 1983, Jones 1984). Differences in width and other features of

daily growth increments have been correlated to life-history transitions, changes in environmental conditions such as temperature and ration size, and physiological factors (for review see Campana & Neilson 1985, Jones 1986).

Flounders have a particularly complex first year because, not only their habitat, but also much of their body form and behavior changes drastically at metamorphosis (35–56 d after hatching). Hatching as symmetrical larvae that feed planktonically, they begin to frequent the bottom as their dorsoventral dimension increases, the notocord tip bends, and the adult-shaped caudal fin develops (Klein-MacPhee 1978). Finally, the fish spends less time swimming in the water column and becomes benthic. This occurs as one eye migrates across the dorsum to the opposite side of the head and the juvenile flounder orients at a 90° angle to its previous alignment. Otoliths do not change their position in the head during this transformation (Platt 1973, Policansky 1982), so the sagittae end up lying one over the other. Evidence of the fish's orientational change may be reflected in the otolith depositional pattern.

Furthermore, physiological changes as well as the change in food source might also register on the otolith.

This study is the first in a continuing sequence of investigations on the effects of environment and physiology on the formation of subannual increments in winter flounder otoliths. Relationships between behavioral and anatomical changes were defined and correlated with baseline data on daily growth increments. Growth rates of both wild and laboratory-reared larvae were determined. Increment counts and morphological changes from known-age laboratory-reared fish were compared with those of wild fish to establish anatomical markers in the otolith during the first year.
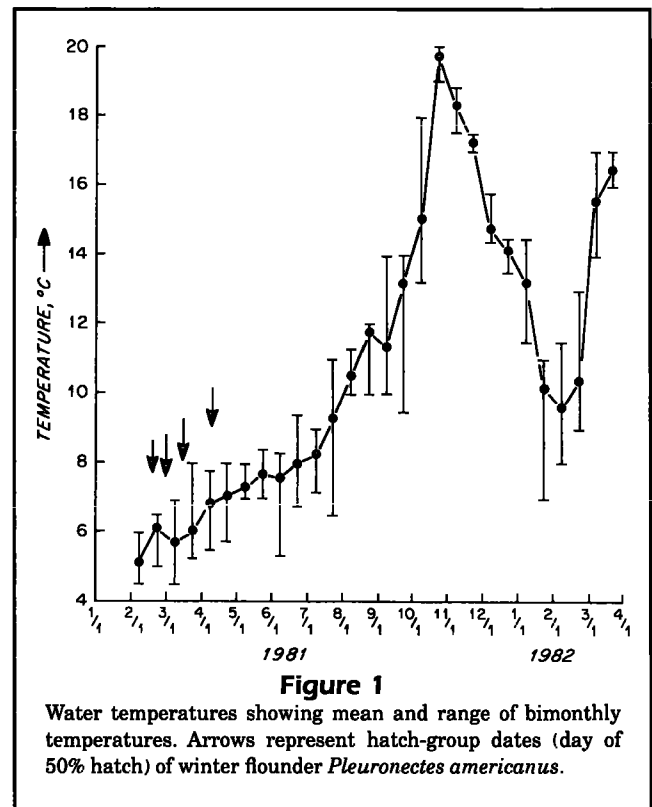
Materials and methods

Acquisition of eggs and larval rearing

Adults caught in Narragansett Bay served as gamete sources. Eggs fertilized in the laboratory were acquired from the National Marine Fisheries Service (NMFS) and Environmental Protection Agency (EPA) laboratories at Narragansett, Rhode Island during February and March 1981. Larvae hatched 15 March 1981 (termed the Mar 15 group) were reared in static trays using methods of Klein-MacPhee et al. (1980). The light cycle was maintained at 11:13 (light/dark). Light intensity in the growth trays varied between 256 and 1777 lux. A separate 114 L tank was maintained for behavioral observations. Light intensity in this tank was 847–4780 lux. Salinity range was 32–33 ‰, temperature was kept at 5–10°C (±1°C) without diel variation until July, when temperature was allowed to roughly follow seasonal patterns (Fig. 1). Larvae were fed once daily unicellular green algae *Tetraselmis souscii* and rotifers *Brachionus sp.*, beginning at 3–5 d before yolksac absorption began. Rotifer concentrations of at least 1/mL were maintained, although concentrations ranged to over 20/mL. When larvae were 40 d old, newly-hatched brine shrimp were added to maintain prey concentrations of 1/mL. After several months, fish were fed frozen brine shrimp with the addition of chopped mussels at irregular intervals.

Heavy mortalities (>90%) reduced larval populations such that only 30 fish survived through metamorphosis (40–60 d posthatch). Of the metamorphosed fish, 11 lived 1 yr and then were killed for otolith examination. Due to physiological effects induced by natural mortality, only otoliths from sacrificed fish were examined.

Sampling

Initially, 10 fish per day selected at random were removed and preserved in 95% ethanol. After hatching,

Figure 1
Water temperatures showing mean and range of bimonthly temperatures. Arrows represent hatch-group dates (day of 50% hatch) of winter flounder *Pleuronectes americanus*.

fish were sampled at 1–6 d intervals until metamorphosis. In an effort to conserve the 30 fish surviving metamorphosis, additional samples of laboratory-reared larvae in 95% ethanol were provided for ageing by the NMFS Narragansett Laboratory (hereafter referred to as the LR hatch group). These samples were of larvae 0–55 d old and were reared using the same methods as the Mar 15 hatch group.

Standard length measurements were obtained from preserved fish. To check for shrinkage in body length, larvae were measured before and after 1 week's preservation and percent shrinkage determined.

Collections

Wild young-of-the-year (YOY) winter flounder were collected throughout summer 1981 using beach seines from estuaries along Cape Cod. Otoliths were dissected from these specimens.

Otolith preparation and analyses

Body lengths of preserved larvae were measured on glass slides. In larvae (hatch to ~30 d), sagittae could not be distinguished from the asterisci or lapilli; therefore all otolith pairs were removed and aged. In fish
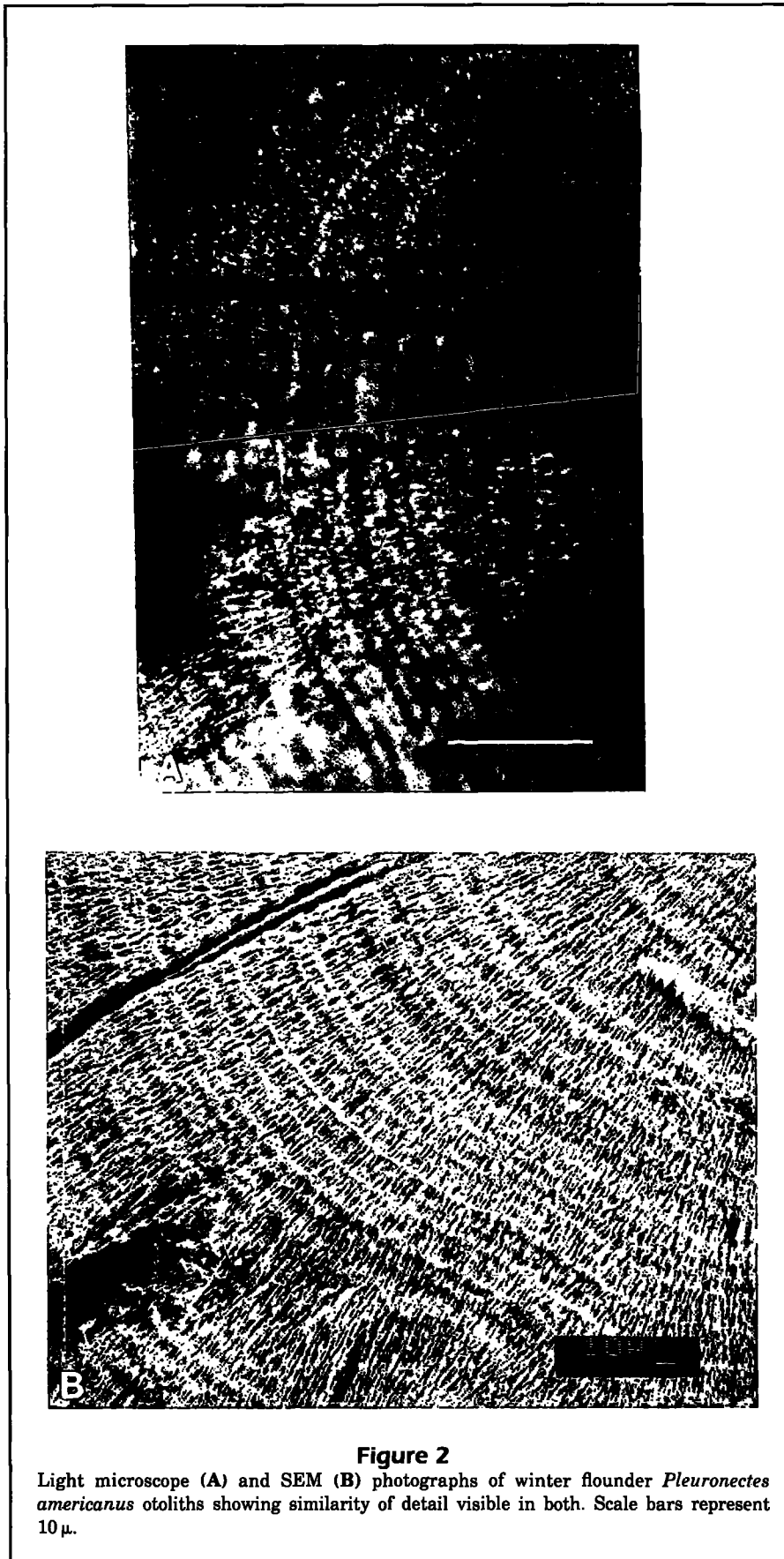
Figure 2
Light microscope (A) and SEM (B) photographs of winter flounder *Pleuronectes americanus* otoliths showing similarity of detail visible in both. Scale bars represent 10 μ.

30 d and older, sagittae were measured and used for increment counts.

Otoliths were measured to the nearest micron (under a compound microscope) along the longest axis through the central core and along the axis perpendicular to that dimension using an optical micrometer at 200–1000× (depending on otolith size). Most increment counts were done on photographs at 1000×. All increments visible in at least two places on an otolith were counted. Varying the focus changed the resolution of increments; therefore the maximum number of increments seen in a series of pictures taken at slightly-varying focal planes was counted. Two or three separate counts by two age-readers were averaged. If the two readers disagreed by more than two increments or the photographs were considered unclear, that otolith set was not used in daily growth-increment calculations. Increments formed prior to yolksac absorption were either absent or difficult to resolve and were not included in the total count. Based on the work of Radtke & Scherer (1982), a correction factor of 10 was added to the number of observed increments in order to establish each larva's estimated age in days from hatch.

For scanning electron microscopy (SEM) viewing, some of the larger (600–840 μ along the largest dimension) sagittae were prepared according to the methods in Radtke & Dean (1982). Light microscope pictures of an otolith were compared with SEM photographs of the same specimen (Fig. 2) and counts were found to be comparable.

Behavioral observations

Larval behavior was observed from hatching through metamor-

phosis to correlate behavior with physiological (as registered in otolith development) and morphological changes. Observations of the larvae were made in holding tanks throughout the period they were reared. Individual larvae were also observed in small containers under low magnification to verify anatomical changes as well as details of small movements.

On the 48th day after hatching, individuals were moved into an observation tank to facilitate observation. An undergravel filter bed was placed in the refrigerated observation tank to minimize disturbance resulting from maintenance procedures. Temperature, diets, and light-cycle conditions were the same as those for separate tray-reared larvae. Light intensity was higher in the observation tank than in trays because overhead lights were supplemented with tank lights. Fish were observed for 10 min periods twice daily, at 10 a.m. and 4 p.m. One fish chosen at random was followed as long as it could be seen; if it moved out of sight, another individual was selected for the remainder of the observation period. Behaviors recorded included swimming (duration, vertical and horizontal direction, body orientation in relation to the bottom, and fin usage); feeding, both before and after adding food (frequency, location, sequence of body motions, success); resting (duration, location, body position); and interactions between individuals. Observations were terminated several weeks after fish had metamorphosed and behavior patterns had stabilized (i.e., assumed a typical adult sedentary behavior pattern).

Figure 3

Gompertz growth curve and equation fitted to plot of standard length and estimated age in days (no. of otolith increments + 10) for 113 larval winter flounder *Pleuronectes americanus*.

Results and discussion

Larval growth rates

From analysis of 113 preserved larval winter flounder ranging from 2.5 to 9.0 mmSL, growth was best described by a Gompertz-type curve (Fig. 3). Previous uses of the Gompertz growth curve and methodology for fitting the curve are described in Pennington (1979) and Bolz & Lough (1988). The variance was stabilized by using the natural log form of the growth equation, and parameters were derived by nonlinear estimation techniques resulting in the relationship:
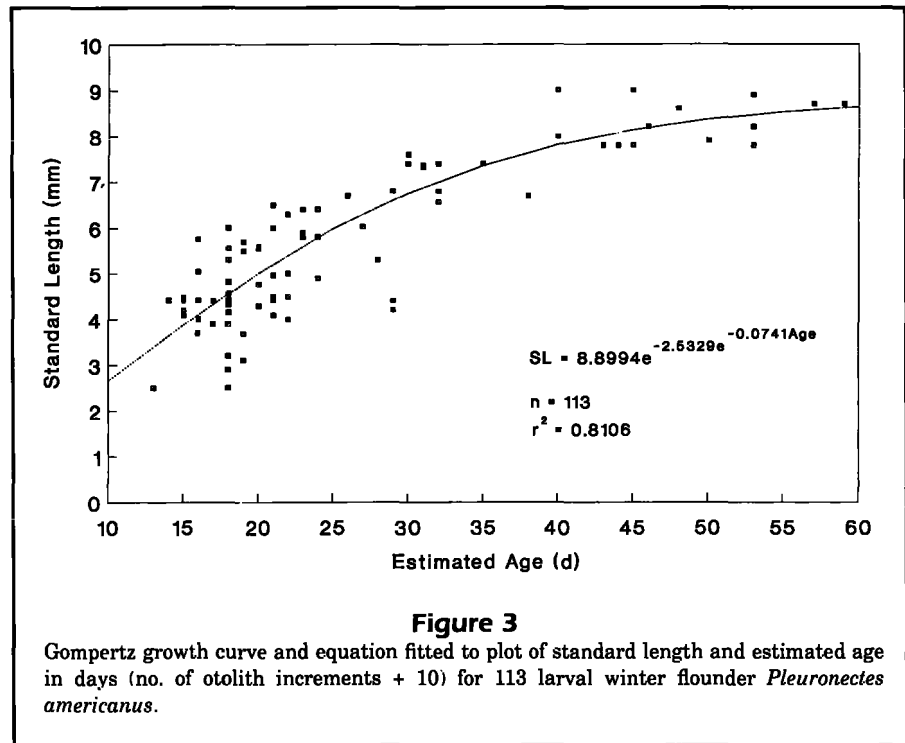
$$\ln(L) = -0.3469 + 2.5329(1 - e^{-0.0741R}), \quad r^2 = 0.8106, \quad (1)$$

where L = standard length in mm, and R = estimated age (increments +10) in days.

The predicted length of 2.66 mm at yolksac absorption compares favorably with that found by Radtke & Scherer (1982) for wild larvae (2.5 mm). The asymptotic length of 8.9 mm probably delineates mean length at metamorphosis and falls within the range (7–13 mm) given by Fahay (1983). The average growth rate (from Eq. 1) for the period under study was 0.31 mm/d, which is slightly less than that observed in the 1982 study using preserved lengths by Radtke & Scherer (0.38 mm/d).

Shrinkage

Little shrinkage was observed for larvae 4–35 d old as has been reported by other researchers (Radtke & Waiwood 1980, Theilacker 1980). Fresh lengths were 2.8–5.0 mm; preserved lengths, 2.5–5.0 mm (n=28) with average shrinkage of 4.2% (SE=0.6). For 91–112 d old flounder, fresh lengths were 5.9–13.8 mm; preserved, 5.8–13.6 mm (n=19) with average shrinkage of 8.6% (SE=0.9). Radtke & Scherer (1982) found no shrinkage in small larvae (<4.7 mm) and only minimal shrinkage (4%) in older fish. Though we observed slightly larger shrinkage than Radtke & Scherer (1982), the growth rates during the 55 d agree roughly with larval flounder growth rates found in their study.
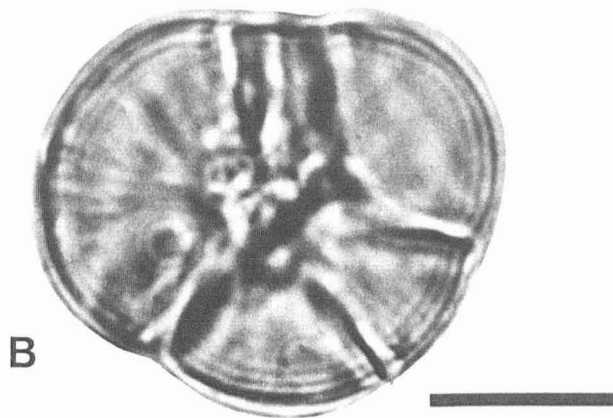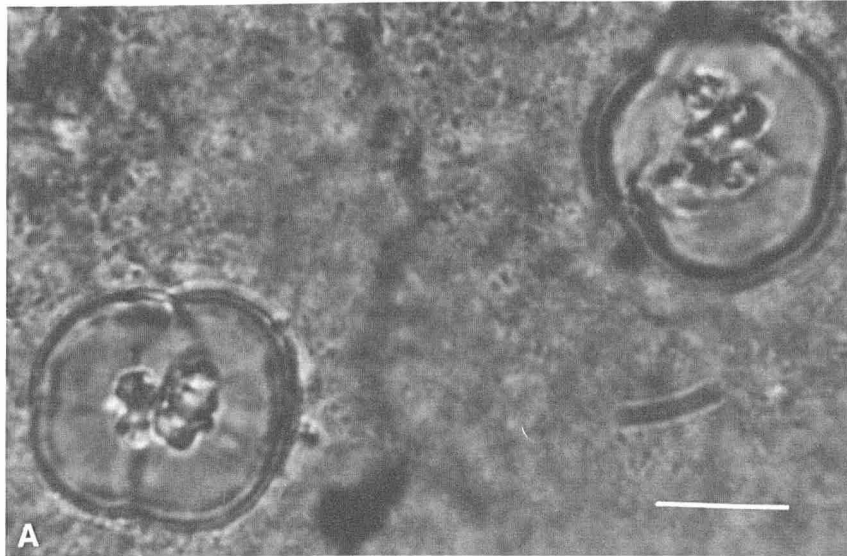
Figure 4
(A) Otoliths taken from winter flounder *Pleuronectes americanus* embryo 13 d after spawning, showing primordial granules in center. **(B)** Rings in otolith taken from embryo 16 d after spawning. Magnification 1000×.

Otolith development

Pre-hatch formation All three pairs of otoliths were present in embryos as early as 13 d after spawning. Premordial granules of material were evident at this time, clumped together in the otolith core (Fig. 4A). The periphery of the otolith forms a rather irregular sphere. Up to four growth rings could be seen on some embryonic otoliths (Fig. 4B). Similar formations have been found on embryonic otoliths of several other species (Taubert & Coble 1977, Brothers & McFarland 1981, Radtke & Dean 1982, Geffen 1983, Brothers 1984), but their periodicity or significance has not yet been determined.

Shape change at metamorphosis At the time of eye migration (40–50 d posthatch), the sagittae of winter flounder underwent a profound change in shape. Clumps of what seemed to be amorphous calcareous material accumulated at the otolith periphery. These accessory growth centers developed irregularly, sometimes appearing two or three on an otolith, often forming at 90° intervals around the circumference of the previously round otolith (Fig. 5A). Similar observations have been made in other Pleuronectids (Brothers 1984, Campana 1984c).

It is significant that accessory growth centers were found only on otoliths of flounders during and after metamorphosis. Larvae with symmetrically-placed eyes did not show these irregular formations on their sagittae, even as late as 73 and 76 d posthatch (Fig. 5B). A photograph of a non-metamorphosing 73 d-old larval otolith without accessory growth centers is compared with that of the typical otolith from a metamorphosing individual in Figure 5. The appearance of asymmetrical formations on sagittae of metamorphosing flounders coincides with the change from vertical to horizontal orientation (i.e., dorsal-side uppermost to right-side uppermost) which accompanies the shift to an asymmetrical form and a benthic habitat. Thereafter, accretion again seems to proceed by increments which coincide with age in days, but which continue an asymmetrical deposition until the adult shape is stabilized. If the formation of accessory growth centers is found to occur at the time of metamorphosis in other flounder species (Brothers 1984, Campana 1984c), this may prove useful in marking the point of habitat change within the otolith record. It is possible that accurate otolith counts could then begin with the juvenile stage rather than the earlier, less easily prepared, and counted larval otoliths.

Fish length/otolith length relationship during the first year The relationship between sagitta size (larg-
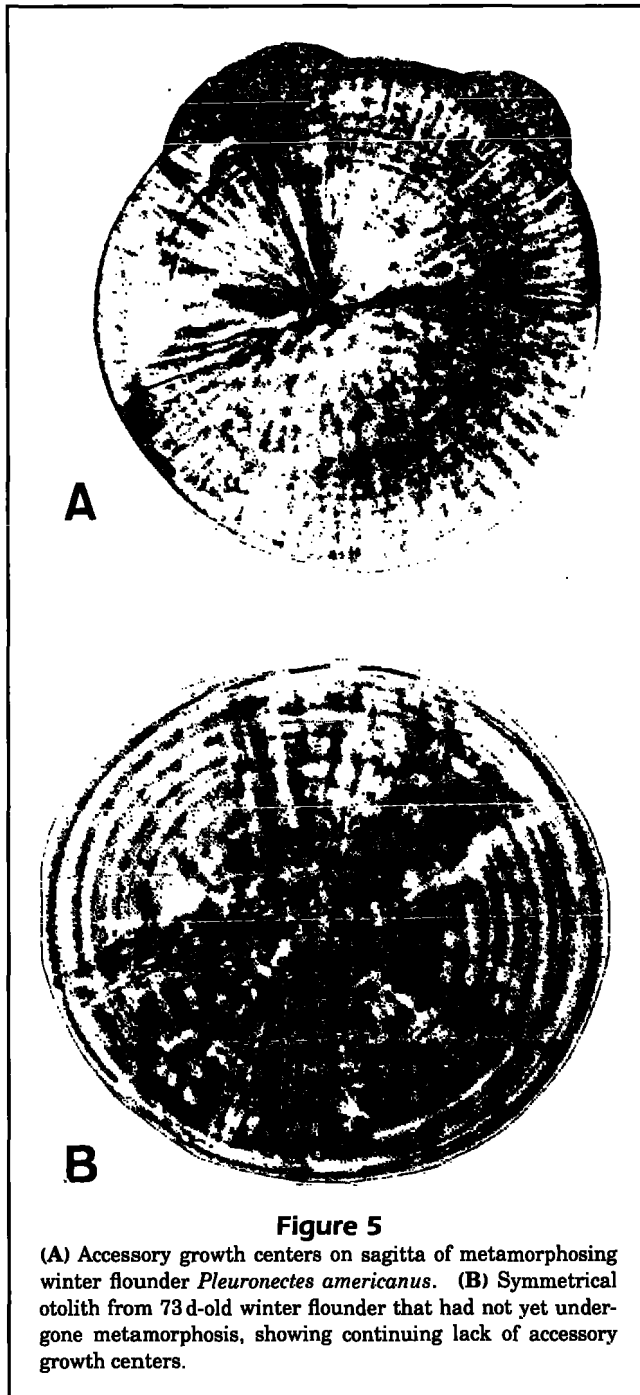
Figure 5
(A) Accessory growth centers on sagitta of metamorphosing winter flounder *Pleuronectes americanus*. (B) Symmetrical otolith from 73 d-old winter flounder that had not yet undergone metamorphosis, showing continuing lack of accessory growth centers.

est dimension) and fish length was nonlinear for premetamorphic larvae raised under laboratory conditions (Fig. 6). The best fit equation was exponential,

$$Y = 7.8e^{0.3x} \quad (r^2=0.87),$$

where x is standard length and Y is otolith length. Because larger larvae had an average shrinkage of 8.6% as compared with 4.2% for smaller larvae, the

parameter estimates in the above equation may not be bias-free. By the end of the first year, however, the relationship was linear (Fig. 7). Covariate analysis indicated that the regression lines for laboratory-reared and wild YOY winter flounders were not significantly different, so data pairs were pooled. The resulting regression line for YOY flounders (about 3 mo or older) using the same variables as above was
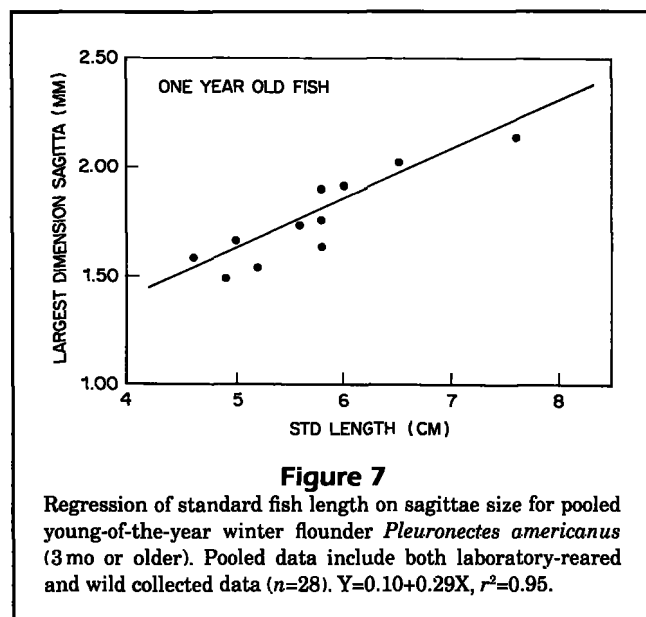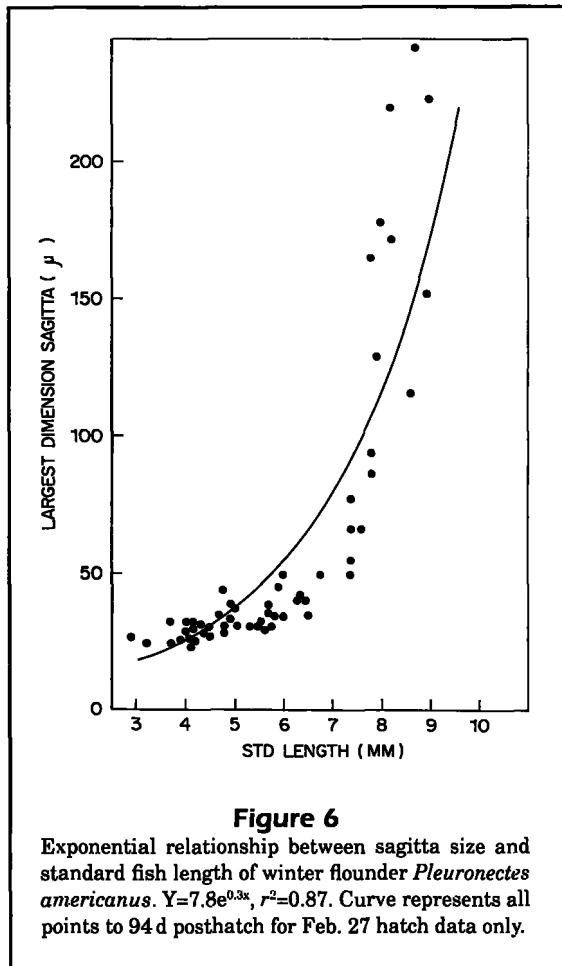
$$Y = 0.10+0.29x \quad (r^2=0.95).$$

Both linear and allometric relationships between otolith size and larval fish length have been reported in the literature (Taubert & Coble 1977, Brothers & McFarland 1981, Methot 1981, Radtke & Dean 1981). Although this relationship has been reported for starry flounder (Campana 1984c), it has not been previously reported for larval winter flounder. It is not surprising, however, that otolith growth exceeds growth in body length. Addition to body depth is enhanced as the body form alters towards the adult shape. This feature may compensate for the decline in growth in length at this time (Pearcy 1962, Laurence 1975). Investigating the relationships between length, depth, otolith dimension, otolith mass, and fish mass may in future studies elucidate the relationship between larval flounder somatic growth and otolith growth.

Wild vs. laboratory-reared fish Otoliths from wild juvenile samples showed the same diameter/fish length relationship as otoliths from our laboratory-reared fish. Otoliths from wild fish exhibited a more regular and somewhat sharper depositional pattern of increments. Therefore, these were used more frequently for SEM analysis. The superior clarity and regularity of otolith incremental patterns from wild fish over laboratory-reared fish have been discussed in the literature (Blaxter 1975, Uchiyama & Struhsaker 1981, Radtke & Dean 1982, Radtke & Scherer 1982) and is discussed in detail by Campana & Neilson (1985).

Early larvae: Hatch to 40 d Under a temperature regime of 5–7°C, larvae hatched 14–18 d after being spawned. Hatching was accompanied by intermittent writhing and vibrating motions of the embryos. Hatchlings sank to the tray bottom when not in motion.

Swimming began immediately after hatching. Larvae swam with rapid lateral tail-whips and could control direction. All swam away from disturbances caused by a pipette tip. Swimming became stronger and more sustained over the first 10 d posthatch. On the third day after hatching, a series of brief (1–8 s) upward swims, followed by a 20 s to 1 min passive period resulting in head-down sinkings, were first noted. This intermittent swimming behavior may be adaptive in

Figure 6

Exponential relationship between sagitta size and standard fish length of winter flounder *Pleuronectes americanus*. $Y=7.8e^{0.3x}$, $r^2=0.87$. Curve represents all points to 94 d posthatch for Feb. 27 hatch data only.

Figure 7

Regression of standard fish length on sagittae size for pooled young-of-the-year winter flounder *Pleuronectes americanus* (3 mo or older). Pooled data include both laboratory-reared and wild collected data ($n=28$). $Y=0.10+0.29X$, $r^2=0.95$.

protecting young larvae from extensive transport by surface currents (Sullivan 1915, Pearcy 1962). Sullivan reported that swimming appeared to be periodically inhibited by a factor other than fatigue. By 7 d, larvae swam for 3–10 s followed by a 2–10 s passive period. After about 25–30 d, swimming was constant during the day except when interrupted by feeding behaviors.
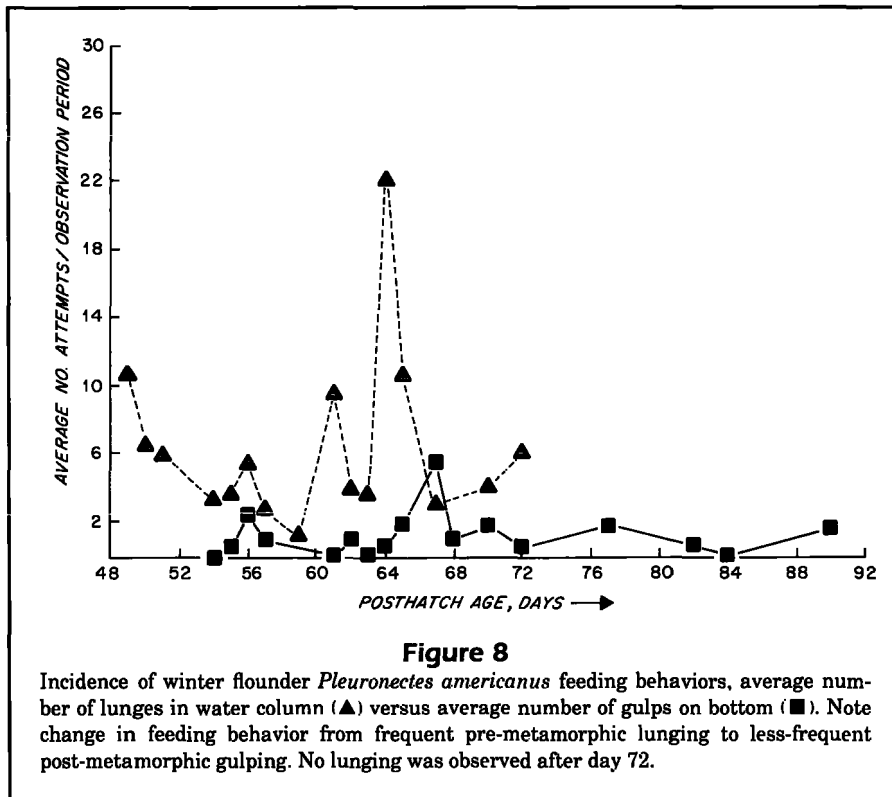
For the first 10 d posthatch, swimming larvae were concentrated at the surface; after about 20 d, larvae were dispersed throughout the water column. There were noticeable aggregations of larvae at tank corners and along sides, but no schooling behavior was observed. Swimming larvae avoided bright microscope lights.

Lunging—defined as a sudden, rapid thrusting motion of the body which propels the larva less than a centimeter forward but which is faster and more abrupt than swimming activity—was observed as early as 5–7 d posthatch (depending on hatch group), before the mouth was completely formed. Once the mouth parts had formed (at the time of yolksac absorption), lunges included rapid and wide jaw gape and snap. Incidence of lunging increased from less than once a minute initially to once every 10 s or more frequently by 40 d posthatch. Systematic observations of lunges began at day 48 (Fig. 8). Prey items were not always visible but were seen often enough that these movements were assumed to be feeding lunges. No lunges were observed after day 72 coincident with metamorphosis.

A sigmoid coiling of the body, called an "S motion" here, often preceded the lunge. This motion could be slow or fast, and when rapid often included a single side lunge as the body was pulled backwards and the head whipped from one side of the "S" to the other. The rapid "S" was first observed 5–9 d posthatch, while the slow one was not noted until 30–50 d posthatch. Larval feeding by such an "S strike" motion has been described for other species in the literature (Rosenthal & Hempel 1970, Hunter 1972). The slow "S motion" we observed in older larvae is speculated to be related to the greater accuracy of striking prey facilitated by experience.

Successful feeding, defined by observation of at least one food particle in the gut of sampled larvae, began at 9–14 d posthatch, at or just after yolksac absorption (8–12 d posthatch). However, growth has been reported to slow or stop for several days after absorption of the yolksac (Cetta & Capuzzo 1982).

Passive, nonswimming yolksac larvae sank in a head-down position in the water column until they hit bottom or abruptly resumed swimming towards the surface. When on the tray bottom, they lay on either side, or on top, of their yolksac. As swimming duration increased, time on the tray bottoms decreased until, after 20 d (when the yolksac was no longer present), few were seen on the bottom. Passive, nonswimming behavior in the water column was, however, observed past 20 d. After yolksac absorption (~12 d posthatch)

Figure 8

Incidence of winter flounder *Pleuronectes americanus* feeding behaviors, average number of lunges in water column (▲) versus average number of gulps on bottom (■). Note change in feeding behavior from frequent pre-metamorphic lunging to less-frequent post-metamorphic gulping. No lunging was observed after day 72.

the larvae began to maintain their bodies in a horizontal position as they sank, instead of sinking in the vertical, passive, nonswimming position.

Behavior at metamorphosis: 40–60 d posthatch Eighteen fish were placed in the observation tank on day 48 so that their behavior could be more closely monitored. The following account is based on those observations.

By 40–50 d posthatch, larval body form began to change. The body widened dorsoventrally, pigment developed (especially over the head, jaw, gut, and fins), and the end of the notocord began to bend as the adult caudal fin formed. These physical changes, described more fully by other researchers (Sullivan 1915, Breder 1922), took place concurrently with the behavioral changes described below.

The most obvious behavioral changes were seen in swimming and resting patterns. Larvae up to 50–60 d posthatch swam upright using the tail-whipping motion. The body was positioned with dorsal fin uppermost, and the eye had not yet migrated. Beginning at 48 d, occasional interruption of swimming was noted as fish drifted in the water column, usually maintaining an upright posture but not moving fins or tail. At 55 d, the first observation of canted swimming was recorded. Fish rose off the bottom in response to disturbances and swam at about a 60° angle to the side, then sank to the bottom again. Only larvae whose eye

was in the process of migration (asymmetrical placement) were seen to swim at an angle in this way, and only infrequently were these individuals observed swimming in the water column.

Fish with obviously widened bodies and adult-shaped tails began the transition to bottom habitat just prior to eye migration (~40–50 d posthatch). Eight fish were observed lying on the bottom by 42 d posthatch, either on their ventral or left sides. Apparently, some of these individuals returned to larval swimming patterns, since only three were not in the water column on day 48. From day 42 until the last record of fish seen in the water column (day 72), swimming fish were seen to sink to the bottom, either head-down or horizontally oriented, and usually rested on their left side for varying periods before swimming up from the sand, again with an upright larval swimming posture (Fig. 9). Other observations of left-side resting have been reported as early as 10–12 d posthatch (Sullivan 1915).

Eye migration was difficult to observe precisely. It has been reported (Sullivan 1915) to occur over an interval of several days, and we observed that it appeared to occur shortly after establishment of the developing larva on the tank bottom. Once eye migration was completed, fish were never seen to swim upright. However, they occasionally entered the water column lying horizontally on the left side, slowly rippling their dorsal and ventral fins rhythmically.

Newly metamorphosed fish were relatively inactive, lying on the bottom for 10 min or more at a time, occasionally moving their eyes. Activity and metabolic levels have been reported elsewhere to decrease dramatically at this point in flounder development (Blaxter & Staines 1971, Laurence 1977). The first slow bottom swimming with rippling dorsal and ventral fins or "creeping" activity was noted on day 55 during metamorphosis.

Bottom-resting fish also "darted" at intervals, swimming in a rapid burst propelled by tail beats. A dart appeared somewhat like a long lunge in its suddenness and in its generation by caudal body motions rather than fin motions. Darting was also observed in unmetamorphosed fish in the water column as an infrequent reaction to a disturbance. The earliest obser-

vation of darting in bottom-resting fish occurred on day 54. Darting persisted as a sporadic activity in metamorphosed juveniles.

Metamorphosing fish fed in the water column as did unmetamorphosed larvae. Feeding behavior was observed throughout the daylight period. Other researchers have noted the strictly diurnal feeding behavior of young winter flounder (Laurence 1977).

Newly metamorphosed fish resting on the bottom were observed "gulping" on day 55. Gulping was a relatively inactive feeding behavior, with the jaw gape and snap found in the lunge but not accompanied by other body movements. Post-metamorphic juveniles increased the incidence of gulps with age, sometimes combining a short creep and a gulp but often showing no other sign of active feeding. After metamorphosis is complete, increased feeding efficiency, coupled with decreased metabolic requirements, results in comparatively low energy expenditures associated with feeding behavior (Blaxter & Staines 1971, Laurence 1977).

Figure 9
Swimming vs. resting behavior of pre-metamorphosed versus developing (eye asymmetrical) or post-metamorphosed winter flounder *Pleuronectes americanus*. 3–18 fish were observed in each 10 min period. Average number observed/period was 7.

Conclusions

In this study, larval winter flounder otoliths were found to reflect internal and external changes indirectly. Daily growth increments did not begin immediately after hatching, although some individuals exhibited otolith rings at hatch. Daily growth increments were not visible beginning at yolksac absorption. Rather, increments were visible beginning at a point midway between yolksac absorption and the beginning of metamorphosis. These increments may have reflected internal changes presaging metamorphosis, although such changes are not yet externally evident. The period during which these increments appeared was also the period during which swimming behavior during the day became constant, except when it was interrupted by feeding behavior. Shortly after this time-period, the slow "S motion" was first exhibited as a feeding behavior, possibly correlated with experience and better feeding efficiency.

Metamorphosis resulted in obvious behavioral changes as well as anatomical ones. Swimming began with an upright position in which the tail was whipped back and forth to provide propulsion. Larvae then went through a period of canted swimming before settling into swimming on their sides using a rippling of their
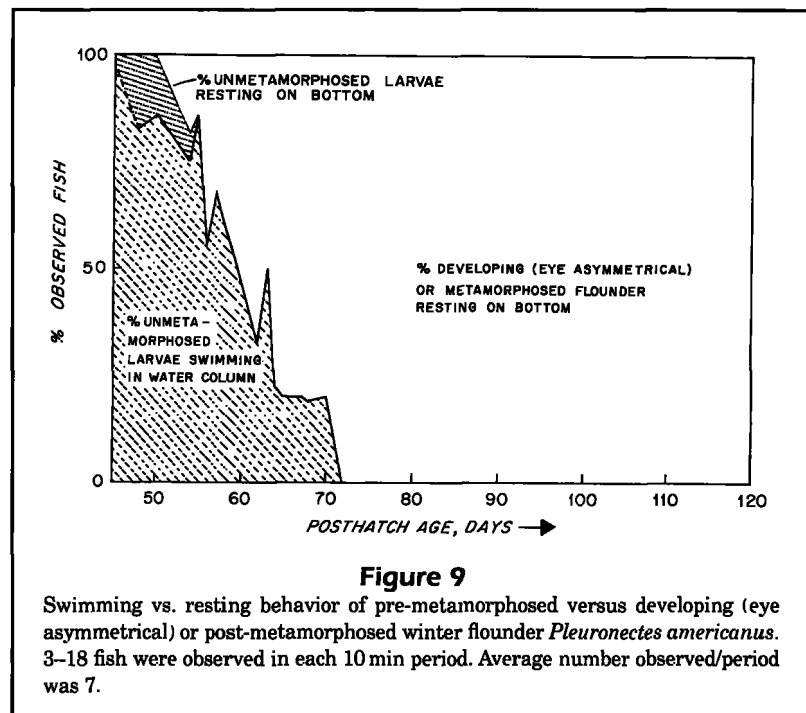
fins for most propulsion. Horizontal swimming with rippling fins was associated with the change to a more benthic existence. The horizontal swimming style occurred less frequently than the earlier upright one, which was consistent with a decrease in overall activity levels. Individuals spent increasing amounts of time resting on the bottom as metamorphosis progressed. At this time, slow bottom "creeping" was first exhibited along with the relatively inactive "gulping" feeding behavior.

The external changes in body shape at metamorphosis corresponded with internal change in sagittae shape. Otoliths from metamorphosed juveniles were found to exhibit accessory growth centers. Sagittae from older fish that had not yet undergone metamorphosis did not exhibit these centers and consequently were still spherical.

The connections between fish growth, behavioral ecology, and physiology are still not well understood. There is clearly a connection between behavior and physiology, although the causality of change in morphology, growth, and behavior is still unclear. This study attempted to correlate some of the behavioral and anatomical changes.

Acknowledgments

Marine Fisheries Service, Woods Hole, Massachusetts. We especially appreciate the aid provided by Louise Dery. We would like to acknowledge the student trainees that made the project possible. Our sincere appreciation to Dr. Werner Graf of Rockefeller University, New York City, who provided his expertise and suggestions. Our thanks also to Dr. Grace Klein-MacPhee, Graduate School of Oceanography, University of Rhode Island, Narrangansett Bay Campus, Kingston, and Alphonse Smigielski, National Marine Fisheries Service, Narrangansett, Rhode Island, for their assistance and suggestions on the proper maintenance of specimens. Finally, we wish to gratefully acknowledge George R. Bolz, Dr. Michael Pennington, NMFS, Woods Hole, and Dr. Bruce Collette, NMFS, National Systematics Laboratory, National Museum of Natural History, Washington, DC, for their reviews and valuable technical assistance.

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