Journal of Fish Biology (1997) **51** (Supplement A), 250–263

Early life history of herring larvae in contrasting feeding environments determined by otolith microstructure analysis

A. FOLKVORD*‡, K. RUKAN*, A. JOHANNESSEN* AND E. MOKSNESS†

**Department of Fisheries and Marine Biology, University of Bergen, N-5020 Bergen, Norway and †Institute of Marine Research, Flødevigen Research Station, N-4817 His, Norway*

Newly hatched autumn-spawned herring larvae *Clupea harengus* were released in two 2500-m^3 outdoor mesocosms and reared over a 2-month period. Hydrographic conditions were similar in the two mesocosms, but the average plankton density was initially more than 10 times higher in mesocosm B compared to mesocosm A ($>1\,l^{-1}$ *v.* $<0{\cdot}1\,l^{-1}$). Half-way through the experiment the feeding conditions reversed with three times higher average densities in mesocosm A than in mesocosm B ($>3\,l^{-1}$ *v.* $\sim 1\,l^{-1}$). Herring larvae were sampled with a 0·3-m^2 two-chambered net twice weekly, and survivors were harvested by draining the mesocosms at the end of the experiment. Otolith growth trajectories of individual larvae were determined by relating radial otolith size with number of increments from the outer edge of the otolith (days before capture). The increment widths during the first 3 weeks after hatching, including the first-check size, were generally wider among larvae from mesocosm B (relatively good initial feeding conditions) than among those from mesocosm A (poor initial feeding conditions). The otolith growth pattern also confirmed that the surviving herring in mesocosm A belonged to the upper size range of larvae in the mesocosm after only 2–3 weeks from hatching; no such trend was found in mesocosm B. In both mesocosms the otolith size-at-age indicated that with the present sampling gear, herring larvae larger than 20–25 mm were underrepresented in the net samples. The information obtained from otolith-size-at-age is compared with other morphometric and biochemical measures of size and condition of larvae obtained throughout the experiment. © 1997 The Fisheries Society of the British Isles

Key words: mesocosm; growth; size-selective mortality; avoidance; starvation; backcalculation.

INTRODUCTION

Otolith microstructure analysis in fish larvae can provide a record of individual growth on a daily basis, and represent a powerful application in ichthyoplankton ecology studies (Campana & Jones, 1992). In a situation where the same population has been sampled more than once over a time period, differences in average individual growth trajectories can, for example, be used to infer size-selective growth and mortality between the sampling periods (Rosenberg & Haugen, 1982). A backcalculation of otolith growth rests on several assumptions, however, and failure to address these could seriously bias the results and their interpretation (Campana & Jones, 1992). Herring *Clupea harengus* L. larvae have been shown to exhibit a daily increment deposition in the sagitta otoliths during semi-natural feeding conditions (Moksness, 1992), and should be well suited for backcalculation studies. One way of avoiding problems relating to the otolith–body size relations, however, is to model the otolith growth itself, rather than the traditional backcalculated somatic growth (Gallego *et al.*, 1996).

Sampling of larvae and juveniles, and the spatial distribution of the population under investigation, present significant challenges in field studies of age and

‡Author to whom correspondence should be addressed. Tel.: +47 55584456; fax: +47 55584450; email: arild.folkvord@ifm.uib.no

0022–1112/97/51A250+14 $25.00/0 © 1997 The Fisheries Society of the British Isles

growth (Butler, 1992). Both large scale environmental gradients, and smaller scale physical processes, add heterogeneity in larval background, and contribute to apparent differences in otolith microstructure between groups of larvae sampled at different periods. Large outdoor mesocosms are considered a suitable means of overcoming problems inherent in field studies. Enclosed and well-defined larval populations can be followed over time under semi-natural conditions to validate otolith growth (Geffen, 1992). Mesocosms also allow some degree of environmental control, thereby facilitating comparisons of larval growth and survival under different conditions (Øiestad, 1990).

Autumn-spawned herring larvae of the same origin were stocked in two similarly sized mesocosms, one with relatively good initial feeding conditions, and the other with marginal initial feeding conditions. The larval otolith size at age was compared between mesocosms, and between larvae sampled at different time periods within mesocosms. The observed otolith growth patterns were related to the population dynamics and the feeding conditions.

MATERIALS AND METHODS

LARVAL MATERIAL AND REARING FACILITY

Herring eggs were obtained from autumn-spawning North Sea herring from the Buchan area. The eggs were incubated at mean temperature (± S.D.) of 13° C (± 0·2) and average 32‰ salinity. About 18 000 larvae, hatched on 1 September, were released 1 day later in mesocosm B. On 3 September, about 5000 1-day-old larvae were released in mesocosm A. Both mesocosms (at Flødevigen Research Station, Norway) had an initial volume of 2500 m^3. The volume in mesocosm B was increased to 4000 m^3 during the experiment. The main experiment lasted until day 65 (mesocosm A) and day 67 after hatching (mesocosm B), at which point the mesocosms were drained and the remaining herring larvae and juveniles were collected. Around 100 live juvenile herring (age 65–67 days) were transferred from each of the mesocosms to holding tanks in the laboratory. These herring were kept at 12° C and were not fed the following 6–7 days.

Water temperature in the mesocosms was measured at least every other day, and salinity and oxygen were measured about once a week. Micro-zooplankton was sampled twice weekly with a 300-l min^{-1} capacity pump. Macro-zooplankton and herring larvae were also collected twice weekly around midnight with a 0·3-m^2 two-chambered net with 500 μm mesh-size. Larvae used for the otolith analysis were stored in 96% ethanol (Moksness *et al.*, 1995). Further details regarding mesocosms, larval material, sampling, and environmental conditions during this experiment are given in Moksness *et al.* (1995) and Folkvord *et al.* (1996) who have reported results from morphometric and RNA/DNA analyses with different subsets of larval material.

LARVAL AND OTOLITH ANALYSES

Larval length was measured under a dissecting microscope to the nearest 0·1 mm before otolith extraction. Both sagittae (when possible) were extracted from individual larvae using dissecting needles. The otoliths were mounted convex side up in clear nail varnish on glass slides. Dry weight of the larva was then measured (60° C, minimum 24 h) on a Sartorius microbalance to the nearest 1 μg. The larval lengths and weights were not corrected for shrinkage (Moksness *et al.*, 1995).

The otoliths were examined using a computerized video system and a light microscope at 1000 × magnification (Andersen & Moksness, 1988). Increments were counted and measured along the longest axis. The first distinctive increment structure outside the core region was termed first-check since neither of the previously used terms, hatch-check (Moksness & Fossum, 1991) and end of yolk sac check (Geffen, 1982) defines accurately the time of check formation (Høie, 1997). Increments outside the first-check measured

0·6–0·8 μm each, and when individual increments could not be resolved clearly in the inner region of the otoliths, average increment widths of about 0·8 μm were used (Andersen & Moksness, 1988). Since some of the inner increments might not be accounted for during the first days after hatching using light microscopy (Campana *et al.*, 1987), the increments were allocated by assigning the outer increment to the day of catch, the penultimate increment to the day before catch, and so on. Otolith-dependent growth rate was estimated by contrasting increment widths at fixed radial distances of 20, 40, 60, 80, 120 and 180 μm from the core. Otoliths were excluded from the analysis in cases where the difference in radii between left and right otoliths exceeded 15%, or the difference in increment count exceeded three increments and 10% of average increment count. Average increment counts and radii were used when data on both otoliths were available, but increment width data were obtained from individual otoliths. One otolith was chosen randomly per larva to maintain the original structure of the raw data and to avoid pseudroreplication by including two data series per larva.

A total of 219 larvae was analysed from mesocosm A, and 179 larvae from mesocosm B. Seventeen of the larvae were excluded based on the above mentioned criteria. Data from both sagittae were available for 335 larvae.

STATISTICAL ANALYSIS

Data were categorized by time of capture in 10-day intervals, including day of termination, and mesocosm unit. Differences in age specific increment widths between capture intervals within each mesocosm were tested by one-way ANOVA. The data were examined for homogeneity of variances (Bartlett's test) and distribution of errors, and Scheffe's *post hoc* test was used following the ANOVA (Sokal & Rohlf, 1981). Differences in increment widths of groups of larvae from different mesocosms were tested by *t*-tests. Increment deposition rate was validated by regressions of increment count *v.* known age (Geffen, 1992). The slope of the relationship was tested against an expected value of one with a *t*-test. A distance-weighted least squares smoothing procedure, with a stiffness factor of 0·25, was used to describe the general growth pattern of increment widths (StatSoft Inc., 1995). Differences between groups were considered significant at probability levels below 0·05. All statistical analyses and data presentations were carried out with Statistica® for Windows (StatSoft Inc., 1995).

RESULTS

Average temperature in both mesocosms declined from about 18° C at release to 10–12° C at the termination of the experiment. The average prey density in mesocosm A was initially low ($<0{\cdot}1\ l^{-1}$), but increased to over 3 prey l^{-1} during the second half of the experiment. In contrast, the average prey density in mesocosm B was initially higher (up to $8\ l^{-1}$) and declined after 1 month to $1\ l^{-1}$ and below (Moksness *et al.*, 1995; Folkvord *et al.*, 1996). These differences in prey density contributed to differences in larval length at age (Fig. 1). The larvae from mesocosm A were generally smaller than those from mesocosm B during the first 40 days after hatching. The herring collected at the termination of the experiment (day 65 and 67 after hatching) averaged 31·0 and 27·4 mm in mesocosms A and B, respectively (*t*-test, $P>0{\cdot}05$). Survival was estimated to be 90% on day 15 after hatching in mesocosm B and 25% in mesocosm A. About 9 and 15% of the larvae were sampled and 17 and 12% survived to the end of the experiment in mesocosms A and B, respectively (Moksness *et al.*, 1995; Folkvord *et al.*, 1996).

The estimated age at first-check formation showed considerable variability, especially in mesocosm A in which estimated age ranged from 0 to 17 days post hatch (Fig. 2). Larvae from mesocosm B exhibited a smaller spread in estimated

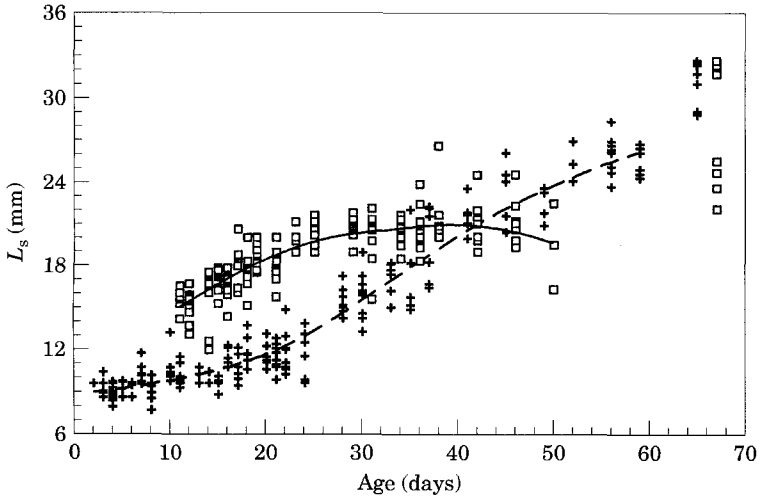

FIG. 1. Standard length (L_s) at age (days after hatching) of individual herring larvae. Distance weighted least squares fitted lines are included to show general trends in growth of net caught larvae in mesocosm A (– –+– –) and B (—□—).

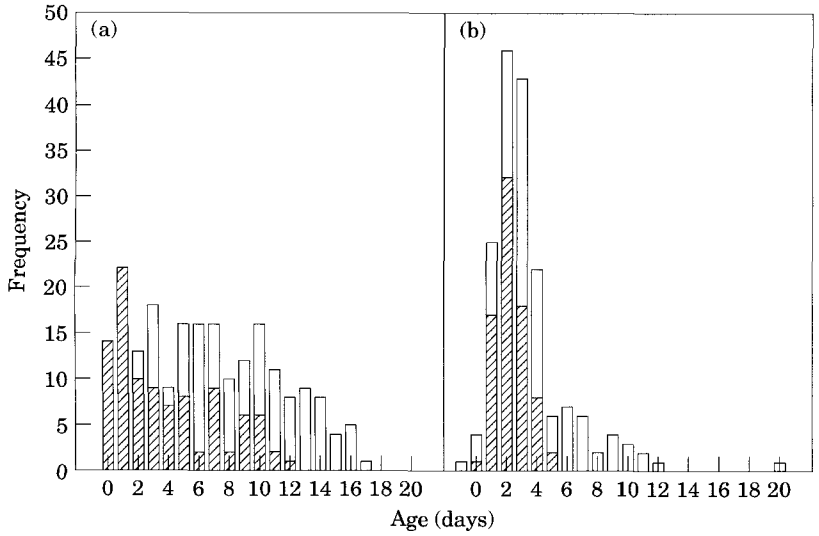

FIG. 2. Frequency of estimated age of first-check formation of larvae from (a) mesocosm A and (b) mesocosm B. The larvae of both mesocosms are grouped into two categories: ▨, day of capture 0–19; □, day of capture 20–67.

age at first-check formation, and for around 80% of the larvae this occurred 2 ± 2 days posthatching. The difference in spread in estimated age at first-check formation in the two mesocosms was most pronounced among the larvae sampled during the first 2–3 weeks after hatching (Fig. 2). The number of visible increments at age thus differed between larvae from the two mesocosms. In mesocosm A, the rate of visible increment formation was clearly <1 day^{-1}

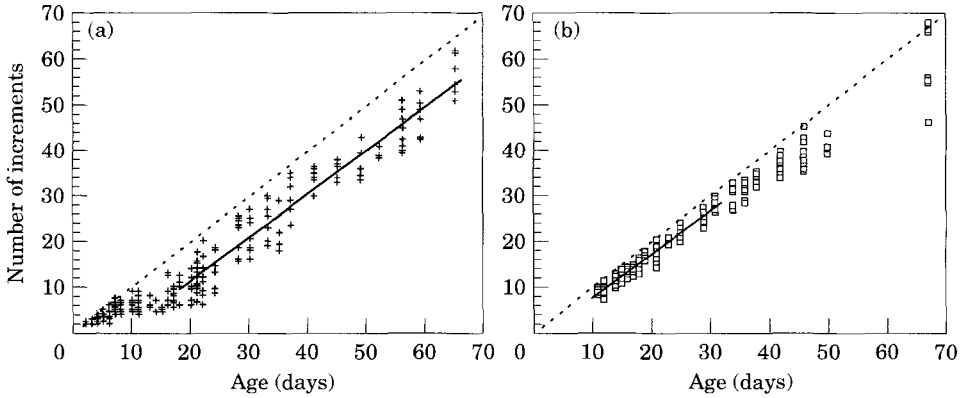

FIG. 3. Number of increments *v.* age (days after hatching) of herring larvae in (a) mesocosm A and (b) mesocosm B. Dashed lines represent $y=x$, and solid lines represent regressions in respective time intervals. (a) $y = -7{\cdot}83+0{\cdot}960x$, (b) $y = -1{\cdot}70+0{\cdot}947x$.

during the first 2·5 weeks (Fig. 3). From day 18, the average increment formation was 0·96 day^{-1}, however, and not significantly different from 1 (*t*-test, $P>0{\cdot}1$). In mesocosm B, the rate of increment formation was <1 day^{-1} during the whole experiment, and averaged 0·89 (*t*-test, $P<0{\cdot}001$). During the first half of the experiment, the average rate of increment formation in mesocosm B was closer to 1 (0·947), but still statistically different from 1 (*t*-test, $P<0{\cdot}01$).

During the first half of the experiment the average age specific increment widths were generally wider in larvae from mesocosm B than A, and during the second month the increments were wider in mesocosm A (Fig. 4; Tables I and II). In mesocosm A, the larvae collected at termination of the experiment also had larger average age specific increment widths than the net sampled larvae from around day 20 and onwards, indicating a size-selective mortality with a subsequent loss of smaller individuals [Fig. 4(a); Table I]. Significant smaller increment widths were also observed at age 20 among larvae caught between days 20–29, compared to those caught between days 40–49 (Table I). Since the larval size in mesocosm A at days 20–29 was below 18 mm, avoidance of the sampling gear was not likely to be a problem at this stage. In mesocosm B, the larvae collected at termination of the experiment had similar increment widths as the net sampled larvae the first 20–30 days of the experiment [Fig. 4(b); Table I]. Thereafter, increment widths of the net-sampled larvae started to decrease whereas the increment widths of the larvae from termination continued to increase for 2–3 more weeks. However, the net-caught larvae sampled during the last time interval before termination in mesocosm B generally had the smallest average increment widths during the whole experiment (Table I).

The otolith size-dependent growth was also generally higher in larvae from mesocosm B than in larvae from mesocosm A, for otolith sizes below 40 μm, with the exception of the slow growing larvae caught before termination in mesocosm B (Fig. 5; Tables III and IV). Larvae from mesocosm B also had a larger average first-check size (10·8 ± 0·7 μm) compared to larvae from mesocosm A (9·7 ± 0·7 μm; *t*-test, $P<0{\cdot}001$) (Table IV). There were slight

TABLE I. Mean otolith increment widths (μm) of larval herring from mesocosms A and B caught in respective time periods at given larval ages

	Time period (days)	Age of comparison (day)					
		10	20*	30	40	50	60
Mesocosm A	10–19	$0{\cdot}85^{a}$					
	20–29	$0{\cdot}71^{a}$	$0{\cdot}89^{c}$				
	30–39	$0{\cdot}75^{a}$	$1{\cdot}08^{bc}$	$1{\cdot}75^{b}$			
	40–49	$0{\cdot}71^{a}$	$1{\cdot}21^{b}$	$1{\cdot}86^{b}$	$3{\cdot}33^{b}$		
	50–59	$0{\cdot}80^{a}$	$1{\cdot}05^{bc}$	$1{\cdot}79^{b}$	$3{\cdot}05^{b}$	$3{\cdot}72^{b}$	
	65	$0{\cdot}80^{a}$	$1{\cdot}61^{a}$	$2{\cdot}63^{a}$	$3{\cdot}97^{a}$	$5{\cdot}23^{a}$	5·41
Mesocosm B	10–19	$1{\cdot}87^{a}$					
	20–29	$1{\cdot}72^{ab}$	$2{\cdot}58^{a}$				
	30–39	$1{\cdot}64^{ab}$	$2{\cdot}53^{a}$	$2{\cdot}06^{b}$			
	40–49	$1{\cdot}46^{b}$	$2{\cdot}26^{ab}$	$2{\cdot}11^{b}$	$1{\cdot}88^{b}$		
	50–59	$0{\cdot}97^{b}$	$1{\cdot}68^{b}$	$1{\cdot}75^{b}$	$1{\cdot}35^{b}$	$1{\cdot}03^{b}$	
	67	$2{\cdot}27^{a}$	$2{\cdot}47^{ab}$	$3{\cdot}09^{a}$	$3{\cdot}83^{a}$	$3{\cdot}66^{a}$	3·74

Mean values with different superscripts (within mesocosm and day of comparison) are significantly different ($P<0{\cdot}05$), with [a] associated with the highest values (Scheffe's *post-hoc* test). *Age where variance was slightly different between groups ($0{\cdot}05<P<0{\cdot}01$).

TABLE II. Results of *t*-tests comparing otolith increment widths of larval herring from mesocosms A and B caught in respective time periods at given larval age

Time period (days)	Age of comparison of larvae from mesocosms A *v.* B (days)					
	10	20	30	40	50	60
10–19	BBB	—	—	—	—	—
20–29	BBB	BBB	—	—	—	—
30–39	BBB	BBB	BB	—	—	—
40–49	BBB	BBB	NS	AAA	—	—
50–59	NS	NS	NS	AAA	AAA	—
Termination	BB	B	NS	NS	AA	AA

NS, no significant difference; letters A and B indicate mesocosm with largest increment widths; one, two, and three letters represent $P<0{\cdot}05$, $P<0{\cdot}01$, and $P<0{\cdot}001$, respectively; —, test does not apply.

differences in first-check size within the respective mesocosms (one-way ANOVA, $P<0{\cdot}05$), but none of the groups was significantly different from the other (Table III). On the other hand, the otoliths from larvae collected at the termination of the experiments had, in general, larger increment widths at any given otolith size compared to otoliths from net-caught larvae in respective mesocosms (Fig. 5; Table III). The larvae collected from mesocosm A at termination had larger increment widths than larvae from mesocosm B during

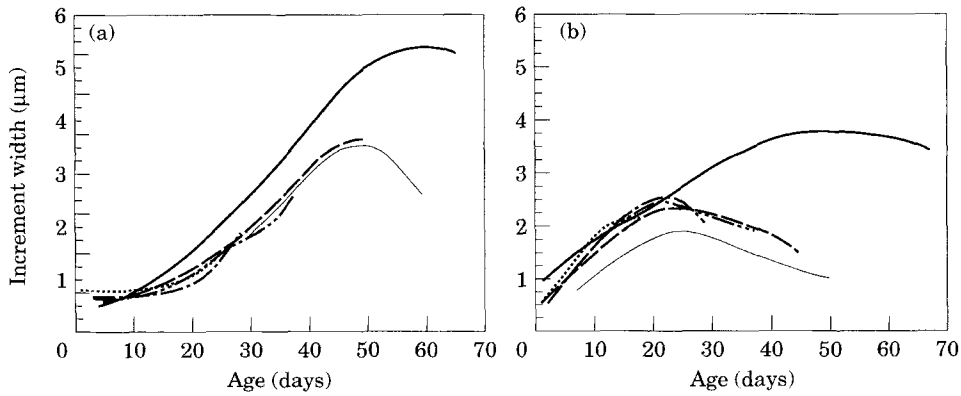

FIG. 4. Age-specific otolith growth of herring larvae in (a) mesocosm A and (b) mesocosm B (increment widths in μm). Lines are estimated by distance weighted least squares fit. Larvae are grouped into categories based on age of capture (days after hatching): · · · ·, 10–19, – · – · –, 20–29; – –, 30–39; – – –, 40–49; ——, 50–59; ——, 65–67.

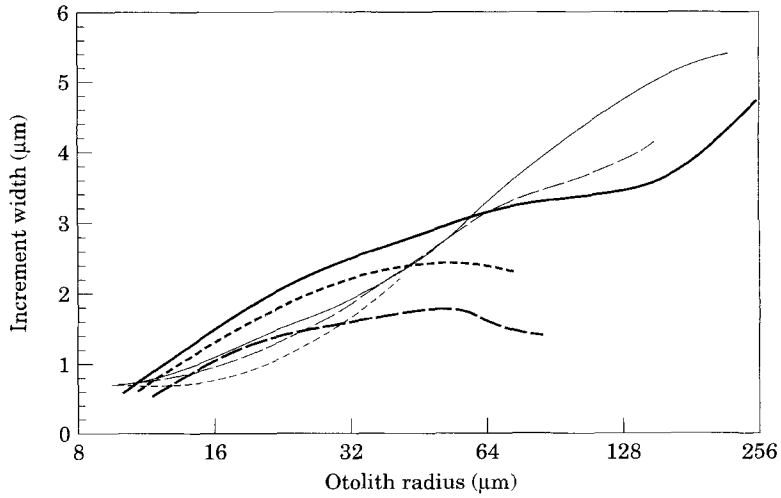

FIG. 5. Otolith size-specific growth of herring larvae in mesocosm A (thin lines) and mesocosm B (bold lines) increment widths in μm. Lines are estimated by distance weighted least squares fit. Larvae are grouped into categories based on age of capture (days after hatching): – – –, 20–29; — — —, 50–59; ——, 65–67.

the last weeks of the experiments, reflecting the better growth conditions in this mesocosm during the last part of the experiment (Table IV).

The larvae transferred to the laboratory after draining mesocosm A, clearly showed a reduction in increment width after transfer to starvation conditions (t-test, $P<0{\cdot}05$; Fig. 6). In mesocosm B, the final increment widths were smaller at initiation of starvation than in mesocosm A (t-test, $P<0{\cdot}01$), and no marked reductions in subsequent increment widths were observed (t-test, $P>0{\cdot}1$; Fig. 6). The average increment widths after 6–7 days of starvation were 3·89 and 2·67 μm in mesocosms A and B, respectively (t-test, $P>0{\cdot}05$).

TABLE III. Mean first-check size (*F*-check) and otolith increment widths (μm) at given otolith radii of larval herring from mesocosms A and B caught in respective time periods

	Time period (days)	Size of comparison (μm)						
		F-check	20	40	60	80*	120	180
Mesocosm A	10–19	$9{\cdot}81^{a}$	$1{\cdot}40^{ab}$					
	20–29	$9{\cdot}46^{a}$	$0{\cdot}99^{b}$	$2{\cdot}20^{a}$				
	30–39	$9{\cdot}67^{a}$	$1{\cdot}18^{b}$	$2{\cdot}17^{a}$	$3{\cdot}17^{a}$			
	40–49	$9{\cdot}74^{a}$	$1{\cdot}25^{ab}$	$2{\cdot}21^{a}$	$3{\cdot}41^{a}$	$3{\cdot}97^{b}$		
	50–59	$10{\cdot}01^{a}$	$1{\cdot}27^{ab}$	$2{\cdot}30^{a}$	$3{\cdot}14^{a}$	$3{\cdot}56^{b}$	$3{\cdot}73^{b}$	
	65	$10{\cdot}06^{a}$	$1{\cdot}57^{a}$	$2{\cdot}51^{a}$	$3{\cdot}23^{a}$	$4{\cdot}37^{a}$	$5{\cdot}17^{a}$	5·80
Mesocosm B	10–19	$10{\cdot}82^{a}$	$1{\cdot}89^{ab}$	$2{\cdot}53^{ab}$				
	20–29	$10{\cdot}91^{a}$	$1{\cdot}80^{ab}$	$2{\cdot}60^{a}$	$2{\cdot}31^{b}$			
	30–39	$10{\cdot}91^{a}$	$1{\cdot}82^{ab}$	$2{\cdot}49^{ab}$	$2{\cdot}11^{b}$	$2{\cdot}60^{b}$		
	40–49	$10{\cdot}56^{a}$	$1{\cdot}82^{ab}$	$2{\cdot}36^{ab}$	$2{\cdot}05^{b}$	$2{\cdot}18^{b}$	$2{\cdot}80^{a}$	
	50–59	$11{\cdot}55^{a}$	$1{\cdot}48^{b}$	$1{\cdot}98^{b}$	$1{\cdot}77^{b}$	$1{\cdot}60^{b}$		
	67	$10{\cdot}26^{a}$	$2{\cdot}04^{a}$	$2{\cdot}59^{ab}$	$3{\cdot}39^{a}$	$3{\cdot}69^{a}$	$3{\cdot}87^{a}$	4·67

Mean values with different superscripts (within mesocosm and compared otolith size) are significantly different ($P<0{\cdot}05$), with [a] associated with the highest values (Scheffe's *post-hoc* test).

*Sizes where variance was slightly different between groups ($0{\cdot}05<P<0{\cdot}01$).

TABLE IV. Results of *t*-tests comparing first-check size (*F*-check) and otolith increment widths at given otolith radii of larval herring from mesocosms A and B caught in respective time periods

Time period (days)	Size of comparison of larvae from mesocosms A *v.* B (μm)						
	F-check	20	40	60	80	120	180
10–19	BBB	NS	—	—	—	—	—
20–29	BBB	BBB	NS	—	—	—	—
30–39	BBB	BBB	BB	A	—	—	—
40–49	BB	BBB	NS	AAA	AAA	—	—
50–59	BBB	NS	NS	AAA	—	—	—
Termination	NS	BB	NS	NS	A	A	A

NS, no significant difference; letters A and B indicate mesocosm with largest increment widths; one, two, and three letters represent $P<0{\cdot}05$, $P<0{\cdot}01$, and $P<0{\cdot}001$, respectively; —, test does not apply.

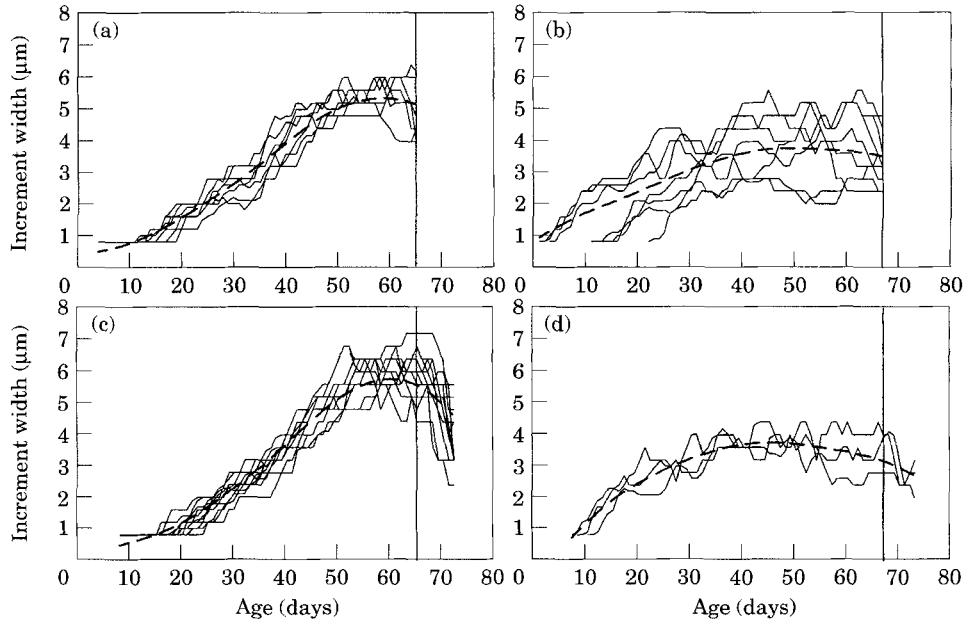

FIG. 6. Individual otolith growth trajectories of larvae at termination of the experiments in (a) mesocosm A and (b) mesocosm B (upper panels), and after 6–7 days subsequent starvation (lower panels). Vertical lines indicate time of terminating the mesocosms. Distance weighted least square fitted lines (– – –) are included to indicate general trends in otolith growth.

DISCUSSION

The larvae from mesocosm B, which had the highest initial growth rate, also had the largest first-check size. It has been noted previously that this first prominent check is formed close to the end of the yolk-sac (Geffen, 1982). In this experiment, no visible yolk remains were seen after day 3 due to the high rearing

temperatures. The larvae used in mesocosm B were from the same brood as those from mesocosm A, but hatched 1 day earlier. Larvae hatched within a day were used in respective mesocosms to increase precision in age estimates (Moksness *et al.*, 1995). A recent study in herring showed no significant differences in sagitta size at hatching of larvae hatching on consecutive days, with the exception of a group incubated at 12° C in which average sagitta size was larger on the day following hatching (Høie, 1997). In this case, the larvae from mesocosm A should then have had larger, not smaller increment widths than those in mesocosm B. Thus the observed differences in first-check size between larvae are not likely to be due to different hatching time, nor temperature-mediated metabolic differences, since both groups were kept in well-mixed water masses with similar temperatures (Folkvord *et al.*, 1996). Elevated metabolic rates have been suggested as the reason for corresponding high rate of otolith formation (Mosegaard *et al.*, 1988; Wright, 1991). The larvae in mesocosm A had very poor feeding conditions, and over 70% were characterized as starving or in poor nutritional condition based on the RNA/DNA index by day 10 (Folkvord *et al.*, 1996). The RNA/DNA ratio of larvae from mesocosm A was also markedly lower than in those from larvae in mesocosm B during the first week after release, suggesting a much lower protein growth rate during this period (Buckley, 1984). It is therefore likely that the larger first-check sizes of larvae from mesocosm B are due to higher metabolic rates associated with feeding and growth compared with the siblings in the other mesocosm.

The age-specific otolith growth confirmed the different growth patterns of herring larvae in the two mesocosms. The increment widths of larvae from mesocosm A were initially narrower than those from larvae of mesocosm B, and these results are in accordance with the results obtained from otolith radius information (Moksness *et al.*, 1995) and RNA/DNA analysis (Folkvord *et al.*, 1996) obtained from other larvae in the same experiment. The average increment widths of larvae from mesocosm A equalled those from mesocosm B some time between days 30 and 40. To the extent increment widths reflect somatic growth, this may imply similar average growth rates of larvae in the two mesocosms during this time interval. Otolith radius and other morphometric and biochemical measures indicated similar growth rates of larvae in the two mesocosms around days 19–26 (Folkvord *et al.*, 1996). A delayed response of otolith increment deposition compared to changes in somatic growth rate has been documented in other species (Bradford & Geen, 1992).

The herring larvae from the two mesocosms collected at the termination of the experiment showed a different response to starvation in subsequent otolith increment width formation. At termination, the larvae from mesocosm A had wider increment widths and higher RNA/DNA ratios (Folkvord *et al.*, 1996) than larvae from mesocosm B prior to starvation, and the subsequent drops in increment widths and RNA/DNA ratios were more marked among larvae from mesocosm A. These results indicated that the larvae from mesocosm A experienced higher growth rates prior to starvation than larvae from mesocosm B. The reduction in increment width related to starvation also confirmed that increment formation did not cease after 6–7 days of starvation at this stage, and that the increment widths of these larger larvae were substantially larger than those from the young starving larvae from mesocosm A which had initial

increment widths below 0·8 μm. The increment widths of herring starved 6–7 days were larger than in autumn-spawned herring during the late larval stage (Moksness & Fossum, 1991), and this emphasises the importance of previous otolith growth history in addition to otolith size in increment width formation (Gallego *et al.*, 1996).

A correct estimation of otolith growth from the outer edge of the otolith towards the core region by allocating 1 increment day^{-1} rests on the assumption that ring deposition is daily (Campana & Jones, 1992). If the daily ring formation is less than 1 as suggested for some time periods in this experiment, the apparent daily increment width will be an overestimate since it incorporates more than 1 day's otolith growth. In mesocosm A the estimated rate of increment formation was not different from 1 during the last 7 weeks of the experiment. The lower apparent rate of increment formation during the first 2·5 weeks may have been due to a non-daily rate of formation during this interval (Geffen, 1982), or resolution limitations with a regular light microscope set-up making increments smaller than 0·4–0·5 μm undetectable (Campana *et al.*, 1987). In any case, the age specific otolith growth during the last 7 weeks in mesocosm A should not have been affected by any irregularities in the otolith formation dynamics during the initial weeks.

The larvae sampled at the termination in mesocosm A can be considered to be the winners in this mesocosm. They were significantly larger than the remaining part of the population by day 20, and kept their relative size advantage throughout the experiment. The maintenance of size ranks within a population has been found in turbot larvae and other species (Rosenberg & Haugen, 1982; Chambers & Miller, 1995; Imsland *et al.*, 1996). Although the initial feeding conditions were poorer in mesocosm A than in B, the winners from mesocosm A had growth rates comparable to the slower growing larvae in mesocosm B. If subsequent size-selective mortality operates in slow-growing populations with poor feeding conditions, a group of survivors with relatively high growth rates will result.

In mescosm A, the size distribution of larvae around day 20 consisted of a significant proportion of larvae with very low somatic and otolith growth. Subsequent sampling revealed that the relative proportion of larvae with minimal otolith growth was reduced, suggesting that significant size-selective mortality took place in mesocosm A after day 20. Such size-selective mortality has been reported for larval turbot in a similar mesocosm (Rosenberg & Haugen, 1982), where the intensity of the size-selective mortality was highest around day 7 after hatching, corresponding to the age at which the starvation group died out. In this experiment, the starvation group had died by day 11 (Folkvord *et al.*, 1996), and the majority of the larvae in mesocosm A were lost by day 15. Some of the remaining mortality taking place during the next 2 weeks was size-selective, and may have been due to the loss of larvae that had been only partly successful in their start feeding. Such a delay of starvation related mortality in slow growing populations has been observed in herring (Werner & Blaxter, 1980), cod *Gadus morhua* L., and shad *Alosa sapidissima* Wilson (Otterå, 1993; Johnson & Dropkin, 1995).

In mesocosm B, the larvae net-sampled prior to termination of the experiment, appeared to be comprised of losers, based on their otolith increment width

pattern. However, the rate of increment formation was lower than 1 during parts of the experiment in this mesocosm. Most noticeably, one of the larvae from the last sampling date contained fewer increments than expected. An underestimation of increments in larvae during the latter half of the experiment in mesocosm B would have resulted in narrower daily increment widths during the same period. The net sampled larvae caught prior to termination in mesocosm B, which was comprised of smaller, slower growing individuals, may have had even narrower daily increment widths during the last time interval. On the other hand, the initial feeding conditions in this mesocosm were relatively good, and under such conditions the majority of the population would have exhibited high growth rates (Øiestad & Moksness, 1981; Folkvord *et al.*, 1994). Thus, no major differences in otolith growth were expected during this stage. The estimated age specific otolith growth of the larvae from the first half of the experiment was not seriously affected by the apparent deviation in daily increment formation, since at most, two increments were lacking according to the observed age–increment relation for this period.

In this experiment, the herring larvae attained a size where avoidance of sampling gear became evident. Sampling was carried out during night to minimize avoidance (McGurk, 1992), but after day 25 in mesocosm B the larvae were not sampled representatively. At this stage herring larvae had reached a size exceeding 20 mm, and previous studies have shown avoidance to be an increasing problem at this stage, especially at lower towing speeds (1 m s^{-1} towing speed used in this study) (Brander & Thompson, 1989). The apparent size-selective mortality in mesocosm A between days 20–29 cannot, however, be explained by avoidance problems since the larvae were smaller than 18 mm during this period. Sound detection is important for successful avoidance at night and the change in avoidance capability at a larval size of 25–30 mm may have been due to the filling of the otic bulla which takes place at this stage (Blaxter & Fuiman, 1990).

The differences in otolith growth between larvae from the two mesocosms were also evident when assessed as otolith size-dependent growth. This strengthens the results obtained in the age-dependent otolith growth analysis. The relative differences in increment widths between the two mesocosms were generally smaller when considered as otolith size-dependent increment widths than as age-dependent increment widths. This suggests that increment widths, at least in part, are related to the otolith size at deposition.

This study has demonstrated the potential use of mesocosms in otolith growth history studies (Geffen, 1992). The virtue of repeatedly sampling an enclosed population of known age is noticeable compared to field based studies. Ultimately, however, there is a need to apply these powerful techniques in the field. Recent field-based investigations (Munk *et al.*, 1991; Gallego *et al.*, 1996; Meekan & Fortier, 1996), using backcalculations of growth in cod and herring larvae and juveniles, have provided new insights into early life-history dynamics of these species under natural settings.

We thank colleagues in Aberdeen, Scotland, for providing us with herring eggs and C. Booman, E. Otterlei, A. Geffen and anonymous referees for constructive comments on a previous version of the manuscript.

References

Andersen, T. & Moksness, E. (1988). Manual for reading daily increments by the use of a computer programme. *Flødevigen Meldinger* **4,** 1–37.

Blaxter, J. H. S. & Fuiman, L. A. (1990). The role of sensory systems of herring larvae in evading predatory fishes. *Journal of the Marine Biological Association of the United Kingdom* **70,** 413–127.

Bradford, M. J. & Geen, G. H. (1992). Growth estimates from otolith increment widths of juvenile chinook salmon (*Oncorhynchus tshawytscha*) reared in changing environments. *Journal of Fish Biology* **41,** 825–832.

Brander, K. & Thompson, A. B. (1989). Diel differences in avoidance of three vertical profile sampling gears by herring larvae. *Journal of Plankton Research* **11,** 775–784.

Buckley, L. J. (1984). RNA-DNA ratio: an index of larval fish growth in the sea. *Marine Biology* **80,** 291–298.

Butler, J. L. (1992). Collection and preservation of material for otolith analysis. In *Otolith Microstructure Examination and Analysis* (Stevenson, D. K. & Campana, S. E., eds), pp. 13–17. *Canadian Special Publications of Fisheries and Aquatic Sciences No.* **117**.

Campana, S. E. & Jones, C. M. (1992). Analysis of otolith microstructure data. In *Otolith Microstructure Examination and Analysis* (Stevenson, D. K. & Campana, S. E., eds), pp. 73–100. *Canadian Special Publications of Fisheries and Aquatic Sciences No.* **117**.

Campana, S. E., Gagne, J. A. & Munro, J. (1987). Otolith microstructure of larval herring (*Clupea harengus*): image or reality? *Canadian Journal of Fisheries and Aquatic Sciences* **44,** 1922–1929.

Chambers, R. C. & Miller, T. J. (1995). Evaluating fish growth by means of otolith increment analysis: special properties of individual-level longitudinal data. In *Recent Developments in Fish Otolith Research* (Secor, D. H., Dean, J. M. & Campana, S. E., eds), pp. 155–174. Columbia: University of South Carolina Press.

Folkvord, A., Øiestad, V. & Kvenseth, P. G. (1994). Growth patterns of three cohorts of Atlantic cod larvae (*Gadus morhua* L.) studied in a macrocosm. *ICES Journal of Marine Science* **51,** 325–336.

Folkvord, A., Ystanes, L., Johannessen, A. & Moksness, E. (1996). RNA : DNA ratios and growth of herring (*Clupea harengus* L.) larvae reared in mesocosms. *Marine Biology* **126,** 591–602.

Gallego, A., Heath, M. R., McKenzie, E. & Cargill, L. H. (1996). Environmentally induced short-term variability in the growth rates of larval herring. *Marine Ecology Progress Series* **137,** 11–23.

Geffen, A. J. (1982). Otolith ring deposition in relation to growth rate in herring (*Clupea harengus*) and turbot (*Scophthalmus maximus*) larvae. *Marine Biology* **71,** 317–326.

Geffen, A. J. (1992). Validation of otolith increment deposition rate. In *Otolith Microstructure Examination and Analysis* (Stevenson, D. K. & Campana, S. E., eds), pp. 101–113. *Canadian Special Publications of Fisheries and Aquatic Sciences No.* **117**.

Høie, H. I. (1997). Maternal, paternal and temperature effects on otolith size of young herring (*Clupea harengus* L.) larvae: an experimental study. Cand. scient. thesis, University of Bergen, Norway.

Imsland, A. K., Sunde, L. M., Folkvord, A. & Stefansson, S. O. (1996). The interaction of temperature and fish size on growth of juvenile turbot. *Journal of Fish Biology* **49,** 926–940.

Johnson, J. H. & Dropkin, D. S. (1995). Effects of prey density and short term food deprivation on the growth and survival of American shad larvae. *Journal of Fish Biology* **46,** 872–879.

McGurk, M. D. (1992). Avoidance of towed plankton nets by herring larvae: a model of night–day catch ratios based on larval length, net speed and mesh width. *Journal of Plankton Research* **14,** 173–181.

Meekan, M. G. & Fortier, L. (1996). Selection for fast growth during the larval life of Atlantic cod *Gadus morhua* on the Scotian Shelf. *Marine Ecology Progress Series* **137,** 25–37.

Moksness, E. (1992). Validation of daily increments in the otolith microstructure of Norwegian spring spawning herring (*Clupea harengus* L.). *ICES Journal of Marine Science* **49,** 231–235.

Moksness, E. & Fossum, P. (1991). Distinguishing spring- and autumn spawned herring larvae (*Clupea harengus* L.) by otolith microstructure. *ICES Journal of Marine Science* **48,** 61–66.

Moksness, E., Rukan, K., Ystanes, L., Folkvord, A. & Johannessen, A. (1995). Comparison of somatic and otolith growth in North Sea herring (*Clupea harengus* L.) larvae; evaluation of growth dynamics in mesocosms. In *Recent Developments in Fish Otolith Research* (Secor, D. H., Dean, J. M. & Campana, S. E., eds), pp. 119–134. Columbia: University of South Carolina Press.

Mosegaard, H., Svedäng, H. & Taberman, K. (1988). Uncoupling of somatic and otolith growth rates in Arctic char (*Salvelinus alpinus*) as an effect of differences in temperature response. *Canadian Journal of Fisheries and Aquatic Sciences* **45,** 1514–1524.

Munk, P., Heath, M. & Skaarup, B. (1991). Regional and seasonal differences in growth of larval North Sea herring (*Clupea harengus* L.) estimated by otolith microstructure analysis. *Continental Shelf Research* **11,** 641–654.

Øiestad, V. (1990). Specific application of meso- and macrocosms for solving problems in fisheries research. In *Enclosed Experimental Ecosystems: A Review and Recommendations* (Lalli, C. M., ed), pp. 136–154. Berlin: Springer Verlag.

Øiestad, V. & Moksness, E. (1981). Study of growth and survival of herring larvae (*Clupea harengus* L.) using plastic bag and concrete basin enclosures. *Rapports et Procès-verbaux des Réunions Conseil international pour l'Exploration de la Mer* **178,** 144–149.

Otterå, H. (1993). Feeding, growth, and survival of Atlantic cod (*Gadus morhua* L.) larvae reared in replicate enclosures. *Canadian Journal of Fisheries and Aquatic Sciences* **50,** 913–924.

Rosenberg, A. A. & Haugen, A. S. (1982). Individual growth and size-selective mortality of larval turbot, *Scophthalmus maximus*, reared in enclosures. *Marine Biology* **72,** 73–77.

Sokal, R. R. & Rohlf, F. J. (1981). *Biometry*. San Franscisco, CA: W. H. Freeman.

StatSoft Inc. (1995). Statistica for Windows (Computer program manual). Tulsa, Oklahoma: StatSoft Inc.

Werner, R. G. & Blaxter, J. H. S. (1980). Growth and survival of larval herring (*Clupea harengus*) in relation to prey density. *Canadian Journal of Fisheries and Aquatic Sciences* **37,** 1063–1069.

Wright, P. J. (1991). The influence of metabolic rate on otolith increment width in Atlantic salmon parr, *Salmo salar* L. *Journal of Fish Biology* **38,** 929–933.