Temperature dependent otolith growth of larval and early juvenile Atlantic cod (*Gadus morhua*)*

E. Otterlei, A. Folkvord, and G. Nyhammer

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The otolith (lapillus) size–fish size relationship was examined for offspring of two Atlantic cod stocks, reared at different temperatures. Larvae and early juveniles reared at high temperatures (fast growing), had larger otoliths at a given length than fish reared at low temperatures (slow growing). Within a given temperature group, however, faster growing cod tended to have proportionally smaller otoliths, although the difference was not always significant. Moreover, the otolith radius of Norwegian coastal cod was larger, at given fish lengths, compared to that of the northeast Arctic cod. An ontogenetic shift in the allometric otolith size–fish size relationship occurred at onset of metamorphosis (12 mm). Mean daily otolith growth of the lapillus radius increased with increasing temperature from 4 to 14°C and was size dependent and peaked at a larval length of about 25 mm. The radius of the lapillus at hatching was poorly correlated with larval length at day 56 for both stocks, suggesting that the potential for fast growth may not necessarily be reflected in traits present at hatching. The effects of temperature, stock and ontogeny are discussed with regard to the assumption of constant proportionality between otolith growth and fish growth.

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Introduction

Given the potential use of the otolith as an individual record of size and growth, it is important to examine the factors that might affect the relationship of otolith growth and somatic growth with regard to the pro-portionality assumption for back-calculation (Hare and Cowen, 1995). The various techniques of back-calculation assume that otolith growth and somatic growth are in constant proportion (e.g. Campana, 1990; Francis, 1990). However, several examples of variable relations between fish growth rate and otolith growth rate have been demonstrated (e.g. Mosegaard *et al.*, 1988; Maillet and Checkley, 1990; Sogard, 1991; Folkvord *et al.*, 1996).

Mosegaard *et al.* (1988) found the response of otolith growth rate to increasing temperature for Arctic char

(*Salvelinus alpinus*) to be totally different from the optimum curve of somatic growth rate, such that otolith growth rate continued to increase at temperatures above those for maximum somatic growth. Faster growing fish also appear to develop smaller otoliths at a given length than slower growing (older) individuals, exposed to the same temperature (e.g. Reznick *et al.*, 1989). A response lag of otolith growth compared with somatic growth, observed for herring (*Clupea harengus*) (Folkvord *et al.*, 1996), and continuing otolith growth of fish during periods of negligible somatic growth (Maillet and Checkley, 1990; Sogard, 1991), implies that there is not a simple proportional relationship between otolith growth and somatic growth on a daily basis (Hare and Cowen, 1995). The functional relationship between oto-lith and fish growth may also differ between ontogenetic stages (Campana, 1984; Hare and Cowen, 1995).

Throughout the extensive area of distribution in the North Atlantic, the several cod (*Gadus morhua*) stocks

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are exposed to a variety of environmental conditions (e.g. Brander, 1995, 1997; Planque and Frédou, 1999), generating systematic differences in ambient temperature between geographic areas. Somatic growth in length and weight of Atlantic cod is significantly influenced by temperature, and stock-specific differences in weight at age are documented (e.g. Brander, 1995; Otterlei et al., 1999). However, the relationship between somatic and otolith growth in cod is more unclear (Geffen, 1995; Miller et al., 1999), and there is little information concerning the relative importance of environmental and genetic factors affecting otolith growth. Here, the main goal was to evaluate the effects of: (i) temperature, (ii) growth rate, (iii) ontogeny and (iv) fish stock on the otolith size–fish size relationship of Atlantic cod larvae and early juveniles and the potential impact on the proportionality of otolith growth and somatic growth.

Materials and methods

A detailed description of the materials and methods used, somatic growth in length and weight including temperature- and stock-specific survival data is presented in Otterlei et al. (1999).

Biological material

Northeast Arctic cod (NA) and Norwegian coastal cod (NC) eggs were naturally spawned during two seasons (5 April 1995 and 14 March 1996) at Parisvatnet, Øygarden and Austevoll Aquaculture Research Stations in western Norway. Eggs were incubated separately in 70 l aerated black conical tanks at 7.3–7.9°C and salinity ranging from 32.9–33.8. In both seasons 50% hatching occurred 12 days after fertilization, referred to as day 0 of larval age.

Experimental design

Two experiments with similar design were carried out in 1995 and 1996. Initial stocking densities were 1400 larvae, 700 NC and 700 NA, per tank. The two-day-old yolk-sac larvae were individually counted and randomly distributed into replicate green, square, fibreglass tanks holding 500 l. In 1995, the fish were co-reared for eight weeks at two different temperatures ($\pm$ s.d.); 4.1 ± 0.2 and 8.0 ± 0.1°C and in 1996 at 6.1 ± 0.1, 10.0 ± 0.2, 12.0 ± 0.3 and 14.1 ± 0.2°C. In order to distinguish NC from NA larvae in 1995, we marked the otoliths of the NA stock with alizarin complexone (100 mg · l^{-1} for 24 h) two days before hatching, whereas in 1996 the otoliths of the NC larvae were marked (Tsukamoto et al., 1989; Blom et al., 1994).

Feeding and rearing conditions

Larvae and juveniles were fed live natural zooplankton in excess (>1000 ind · l^{-1}) and cultivated algae,

Isochrysis galbana and *Rhodomonas baltica*, were added to the rearing tanks. A simulated natural light regime (14L:10D increasing to 19L:5D) for the latitude of Bergen (60°25′N) was used. Temperature was measured twice a day throughout the experiments. The water was gently aerated to reduce the patchiness of the prey and larvae. Oxygen concentration (%) was recorded daily and remained above 70% saturation, while salinity ranged from 30.9 to 33.5.

Sampling procedure

Thirty larvae were routinely sampled weekly from each tank for standard length (SL) measurements. The fish (n=2172) were measured live and transferred individually into marked vials, killed in liquid nitrogen and stored at -80°C for subsequent otolith analysis. The lapilli were extracted under a dissecting microscope equipped with a polarizing filter, and mounted in clear nail varnish on glass slides. Both lapilli were checked for alizarin marks using a fluorescence microscope (Zeiss Axioscope) at $200 \times$ magnification and classified as either NC or NA. Lapilli were selected for a number of reasons (Meekan and Fortier, 1996; Miller et al., 1999), but mainly because it was possible to detect the alizarin mark without further polishing. The right lapillus (left when missing) was examined at 400 to $1000 \times$ magnification, under a fluorescence microscope equipped with a Sony DXC-930P video camera. Maximum radius of the lapillus was measured to the nearest 0.1 µm from the centre of the core to the outer edge of the otolith along the largest axis, and the radius of the hatch check (first prominent D-zone outside the core, Geffen, 1995) was measured from the core along the same axis. Although sometimes difficult to measure, the otolith radius was chosen as an indirect measure of fish size due to its near linear relation to fish length. The choice of reference point or the centre of the core was standardized as follows: (i) when the core consisted of a single granule, the centre of this granule was chosen, (ii) when the core consisted of two smaller granules, the longest radius from the centre of either granule was selected, (iii) when the core consisted of multiple granules, the midpoint of the granules was used as reference point.

The fish alive at the termination of the experiments were counted and sorted with respect to stock. Estimated total survival for the respective treatments was corrected for sampling mortality (i.e., estimated as the mean of the number of fish alive at termination of the experiment and the number of individuals alive at termination plus the number of larvae and juveniles sampled). Survival during the larval stage (SL ≤12 mm) was estimated similarly, corrected for natural mortality recorded during the juvenile stage (for a detailed description see Otterlei et al., 1999).

Data analysis

Initial comparison of larval SL and radius of lapillus was tested with a two-way ANOVA, between stock and year. A nested three-way ANOVA with stock nested in tank, and tank nested in temperature, was further employed to estimate the effects of temperature, stock and tank (replicate) on the mean radius of the lapillus at age. Radius of lapillus did not differ significantly between replicate tanks (nested ANOVAs) and replicates are combined in subsequent graphical presentations. The variables were ln transformed prior to analysis (when needed) to obtain linearity and homogeneous variances (Zar, 1984). Significant ANOVAs were followed by a Tukey's honestly significant difference (HSD) multiple comparisons test to determine differences among treatments.

Otolith size–fish length relationships were evaluated and departures from loglinearity were tested by a second order polynomial fit to the residuals from a common regression of the radius of lapillus versus SL for fish from different stages (Pepin, 1995). Stage-specific differences in otolith size–fish length relations between temperature and stock groups were tested with ANCOVA, using a general linear model (GLM) analysis. Multiple comparisons were employed for common size ranges, larvae (SL$\leq$10 mm, temperatures 4, 6, 8, 10, 12 and 14°C) and juveniles (SL$\geq$14 mm, temperatures 10, 12 and 14°C), with the fish size ranges being determined on the basis of the length-weight relationship previously described in Otterlei *et al.* (1999). The onset of metamorphosis is defined with respect to changes in morphological characters (i.e. the stage where the median larval finfold disappears and is replaced by dorsal and anal fins, a process which is strongly size dependent, occurring at a fish size of 12 mm standard length, and apparently less influenced by age).

Age-independent variability and growth effects on the otolith size versus fish size relationships were estimated for both stocks at the larval and juvenile stages, according to the procedures of Hare and Cowen (1995). The analyses were conducted in several steps, both for multiple temperatures and at temperature-specific levels (at 4 and 14°C for larval NC and at 10 and 14°C for early juveniles from the same stock). First, a preliminary evaluation of somatic size, otolith and age relations was carried out, and the model with the highest correlation, and normally distributed residuals displaying homogeneous variance, was subject to further analysis. Models were kept with as few parameters as possible based on a step-wise regression. Age-independent variability was estimated by calculating the correlation coefficients of the residuals of otolith radius-on-age against the residuals of fish length-on-age (Hare and Cowen, 1995). Age-independent variability is given as the amount of variability in the body size-on-age residuals
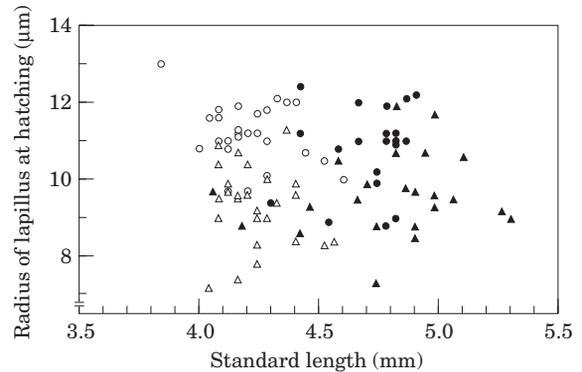

Figure 1. Radius of lapillus (μm) at hatching as a function of larval size in standard length (mm), day 2, for Norwegian coastal cod (NC, circles) and northeast Arctic cod (NA, triangles) used in the experiments in 1995 (open symbols) and 1996 (filled symbols).

that is not explained by otolith size-on-age residuals. Similarly, the effect of growth on the otolith size–fish size relationship was estimated by calculating the correlation coefficients of otolith radius-on-fish length and age-on-fish length residuals (Hare and Cowen, 1995). Throughout the analyses, effects were considered significant at a probability level of α<0.05.

Results

Initial larval SL of a sub-sample (day 2) was not significantly different between the two stocks (two-way ANOVA). The radius of the lapillus at hatching was larger for NC than for NA in both years (12–20%), but not significantly different (two-way ANOVA). Although the mean larval SL of both stocks was higher in 1996 than in 1995 (two-way ANOVA), the hatch check did not differ among years (two-way ANOVA). Moreover, the radius of the lapillus at hatching was poorly correlated with larval SL day 2 for both stocks in 1995 and 1996 (correlation analyses, r<0.32, Figure 1).

Otolith (lapillus) growth

Lapillus size-at-age of larval and early juvenile cod increased successively with increasing temperature from 4 to 14°C (Figure 2). The mean radius of the lapillus generally differed significantly between all temperature groups from day 21 onwards (nested ANOVAs), and already by day 7 all temperature groups differed significantly from adjacent temperature groups except the 8 and 14°C groups. At termination of the experiments the otoliths were 5.7 and 7.9 times larger, respectively, for juveniles of NC and NA reared at 14°C, compared to the otoliths of larvae raised at 4°C. Mean radius-at-age of the lapillus remained significantly larger for the
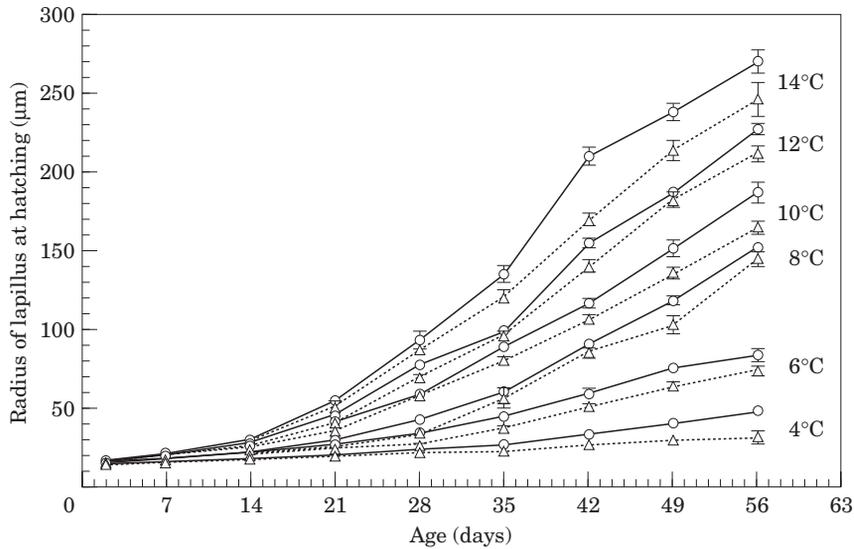
Figure 2. Mean radius of lapillus (μm) at age for Norwegian coastal cod (NC, ○—○) and northeast Arctic cod (NA, △⋯△) larvae and juveniles reared at different temperatures. Vertical lines represent 2 s.e. of the mean.

Table 1. Correlation coefficients of maximum radius of lapillus versus fish standard length and the radius of lapillus hatch check versus standard length, for Norwegian coastal cod (NC) and northeast Arctic cod (NA) larvae and juveniles at termination of the experiments.

Temperature (°C)	Correlation analysis of lapillus maximum radius on length		Correlation analysis of lapillus hatch check on length	
	NC r	NA r	NC r	NA r
4	0.96*	*0.91**	0.03	*0.03*
6	0.80*	0.92*	0.43	0.21
8	0.75*	*0.45*	0.43*	*0.01*
10	0.95*	0.92*	0.19	0.23
12	0.82*	0.71*	0.09	0.10
14	0.87*	*0.52*	0.04	*0.79*

Significant correlations are indicated with asterisk: *. Correlations based on few observations (n=5 to 7) are presented in italics.

NC stock compared to the NA stock throughout the experiments (nested ANOVAs).

As expected, at termination of the experiments, larger cod (in SL) had developed larger otoliths than the smaller ones of the same age reared under identical conditions (Table 1 and Figure 3a–b). However, radius of the lapillus hatch check was generally poorly correlated with larval SL at termination for both stocks (Table 1 and Figure 3c–d).

Effect of ontogeny, otolith radius–fish length relations

A second order polynomial fit to the residuals from the common regression of the radius of lapillus versus standard length for fish from different stages demonstrated departure from log-linearity, with all the parameters significantly differing from zero (nonlinear estimation). The stage-specific otolith radius–fish length relationships of cod indicated positive allometric growth at both the larval and juvenile stages (b>1, Table 2). However, an ontogenetic shift in otolith radius–fish length ratio occurred at the onset of metamorphosis (12 mm SL, Figure 4). Radius of the lapillus of early juveniles (size range 14–45 mm) increased at a greater rate relative to fish length (ANCOVA, b=1.16), than in the larval stage, size range 4–10 mm (ANCOVA, b=1.06, Table 2).

Stage-specific analyses of otolith radius versus fish length for fish of common size are presented using a model without interaction terms (Table 2). The interaction term between stock and temperature made a
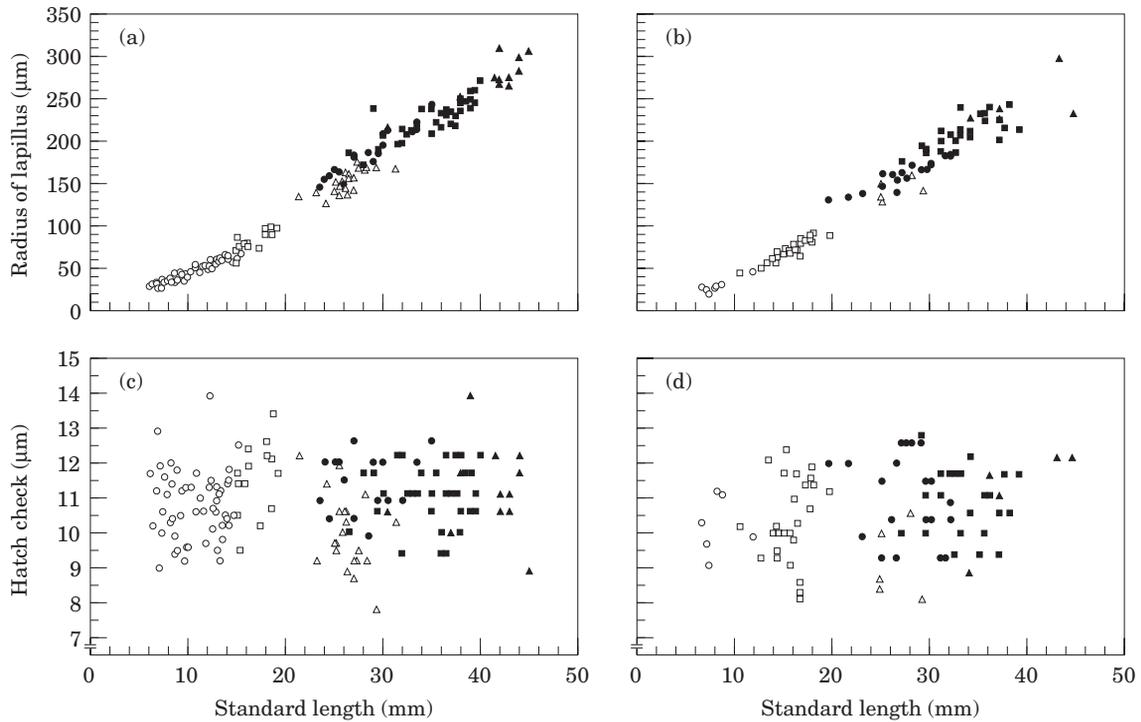
Figure 3. Maximum radius of lapillus (μm) versus standard length (mm) at termination for (a) Norwegian coastal cod (NC) and (b) northeast Arctic cod (NA) larvae and juveniles, reared at different temperatures and radius of hatch check (μm) versus standard length (mm) at termination for (c) Norwegian coastal cod (NC) and (d) northeast Arctic cod (NA) larvae and juveniles from different temperature regimes. Symbols represent 4°C (○), 6°C (□), 8°C (△), 10°C (●), 12°C (■) and 14°C (▲).

Table 2. Temperature-dependent otolith radius–fish length relationships for larvae (SL ≤ 10 mm) and juveniles (SL 14–45 mm) of Norwegian coastal cod (NC) and northeast Arctic cod (NA) of common size ranges.

Stage	Stock	Parameter estimation						b	r²
		a							
		4°C	6°C	8°C	10°C	12°C	14°C		
Larvae	NC	1.190 (261)	1.159 (56)	1.262 (94)	1.250 (38)	1.293 (60)	1.326 (21)	1.064	0.86
	NA	1.135 (152)	1.104 (121)	1.207 (104)	1.195 (73)	1.238 (87)	1.271 (63)		
Juvenile	NC				1.279 (70)	1.301 (145)	1.343 (56)	1.160	0.96
	NA				1.235 (79)	1.257 (120)	1.298 (54)		

The parameters a (intercept) and b (slope) were estimated from the regression equation: $RL = e^a \cdot SL^b$, where RL is radius of lapillus (μm) and SL is fish standard length (mm). Values of the parameter b and determination coefficient r² were common for NC and NA at the larval and juvenile stage, respectively. The number of observations (n) are provided in parentheses.

negligible contribution to explained variance (<0.6%) compared to a more simple model without interactions (larval stage r²=0.86, n=1130; juvenile stage r²=0.96, n=524).

Radius of lapillus at length was generally larger at higher temperatures (ANCOVAs), within both the larval and juvenile stages (Table 2). The lapillus radius of larvae reared at 14°C was 18.1% larger, on average, than
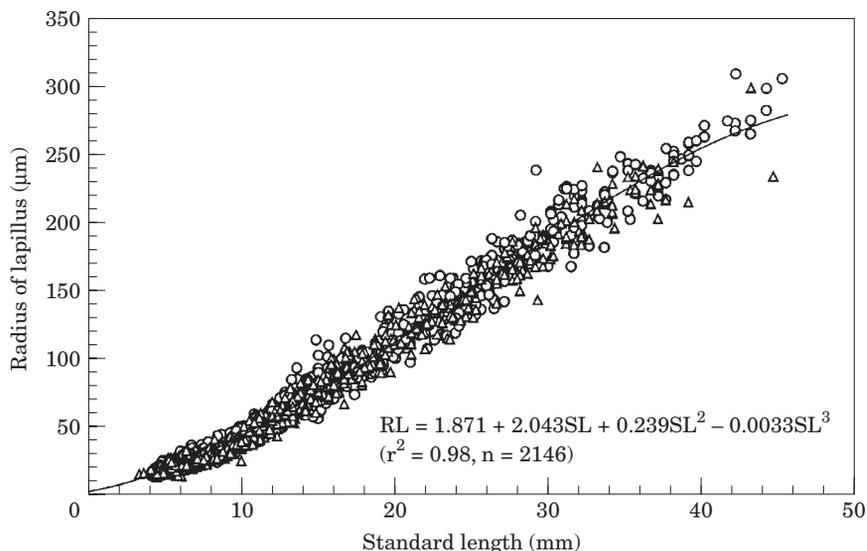
Figure 4. Relationship between radius of lapillus (RL, μm) versus standard length (SL, mm) for Norwegian coastal cod (NC, ○) and northeast Arctic cod (NA, △) larvae and juveniles from multiple temperature groups. A common polynomial of third order is provided.

at 6°C over a common length range (4–10 mm), whereas the otolith radius of juveniles reared at 14°C was 6.5% larger than that of juveniles at 10°C in the length range 14–45 mm. A different otolith size–fish length relation was observed between stocks (ANCOVA). On average, the otolith radius of larval and juvenile NC stock was 5.7% and 4.5% larger, respectively, at given lengths, than the NA stock (Table 2).

Mean otolith growth

Mean radius of the hatch check of lapillus ranged from 10.8–11.3 μm for NC and 9.3–9.6 μm for NA for embryos incubated at 8°C. At day 2, maximum otolith radius was 4.0–5.8 μm larger than the observed hatch check size for larvae reared at the same temperature, indicating a rapid otolith growth around hatching. Pronounced temperature- and size-specific effects on lapillus growth were observed in the experiments (Figure 5a,b). Mean daily otolith growth of the lapillus radius increased with increasing temperature from 4 to 14°C. Maximum (and average) observed otolith growth, at 14°C, for NC and NA were 10.5 (4.6) and 6.9 (4.2) μm d^{-1}, respectively. In contrast, average otolith growth was less than 1.0 (0.4–0.7) μm d^{-1} in both stocks at 4°C. Mean daily otolith growth also increased transiently with increasing fish length, peaking at a juvenile length of about 20–25 mm (Figure 5a,b).

Age-independent variability and growth effects

The correlation coefficients for the residual plots of body length-on-age with the residuals of otolith radius-on-age, including multiple temperature groups of larval cod were 0.84 and 0.80 for NC and NA, respectively (Figure 6a,b), and 0.92 for early juveniles of both stocks (Figure 7a,b). However, the age-independent variability was larger and more distinct when tested temperature specifically. Temperature-specific correlation coefficients for length and otolith radius residuals of larval NC cod were 0.66 at 4°C and 0.87 at 14°C, and 0.90 and 0.85 for early juveniles of the same stock at 10 and 14°C, respectively.

Analysis of age-on-length and otolith radius-on-length residuals of multiple temperature groups demonstrated a weak but significant effect of growth rate on the otolith size–fish size relationship for larval NC cod (r=0.10, Figure 6c). No effect of growth rate was registered for larval NA or early juveniles of the two stocks, (r=0.03–0.10, Figures 6d and Figure 7c,d). However, the effect of growth rate, based on temperature-specific analysis, was significant for larval NC cod at 4°C (r=0.63) and 14°C (r=0.66), and for early juveniles at 10°C (r=0.37) although not significant for juveniles at 14°C (r=0.15).

Survival

Larval stage-specific mortality was high and variable and ranged from 0.035 to 0.109 d^{-1} for NC and from 0.019 to 0.110 d^{-1} for NA. The larval mortality varied with respect to year, temperature and stock, without showing any distinctive trends. Subsequent mortality of fish >12 mm mean SL was low and comprised <2.6% of the initial number of larvae in the tank, indicating no or low cannibalism in the present experiments.
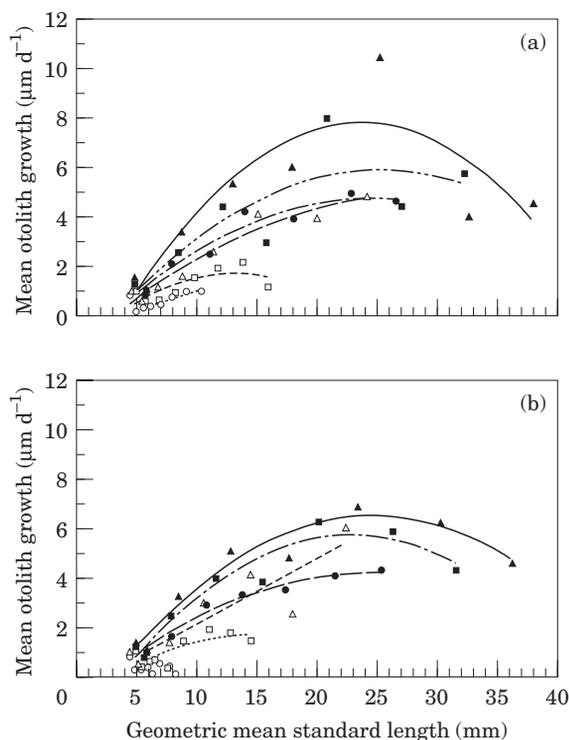
Figure 5. Mean otolith growth (μm d^{-1}) versus geometric mean standard length (mm) in (a) Norwegian coastal cod (NC) and (b) northeast Arctic cod (NA) larvae and juveniles from different temperature regimes. The fitted lines (least square) present the general trends in the otolith growth. Symbols represent 4°C ($\bigcirc$), 6°C ($\square$), 8°C ($\triangle$), 10°C ($\bullet$), 12°C ($\blacksquare$) and 14°C ($\blacktriangle$).

Discussion

Temperature significantly affected lapillus growth in cod larvae and juveniles. The somatic and otolith growth rates increased with increasing temperature over the range 4–14°C for fish fed in excess. Radius of the lapillus was 5.7–7.9 times larger, depending on stock, in early juveniles reared at 14°C compared to fish reared at 4°C, at an age of 56 days. The importance of temperature as a governing factor for otolith growth rate has been well documented (e.g. Mosegaard *et al.*, 1988). For temperatures above optimum for maximum somatic growth, however, otolith formation is expected to behave differently from that of somatic growth (Mosegaard *et al.*, 1988), indicating that the temperature optimum for maximum somatic growth of larval and early juvenile cod is above 14°C, in accordance with Otterlei *et al.* (1999).

As expected, larger cod had larger otoliths than smaller ones of the same age reared under identical conditions. However, radius of the lapillus hatch check was poorly correlated with larval SL at day 56 for both stocks, suggesting that the potential for fast growth may

not necessarily be reflected in otolith size at hatching. Surviving cod in the field had larger hatch checks than the population at large, indicating that the potential for fast growth could be reflected in traits present at hatching (Meekan and Fortier, 1996). This suggestion is not supported by the present results. Recently, Miller *et al.* (1999) suggested that otoliths can, however, be used to infer the standard length of cod larvae at hatching; they argue that variation in egg size rather than variation in larval size at hatching directly may have influenced the conclusions reached by Meekan and Fortier (1996) that there is selection for faster growing larvae. Whether differences in size-dependent mortality or the mortality rate itself can explain these diverging observations between larvae growing up under field conditions versus larvae reared in the laboratory is uncertain. Although the field larvae are survivors of very high mortality, there is no clear evidence whether they only are recruited from the faster growing larvae. The role of size-dependent or -selective mortality in field is still not quite clear.

Despite the overall strong fit to the loglinear relationship between the radius of the lapillus and standard length ($r^2=0.98$, n=2146), the analyses of the residuals clearly show a nonlinear pattern. Reviewing the literature, it has been common practice to describe the relationship between otolith (lapillus and sagitta) size and fish length of both the larval and early juvenile stages of cod, by a single linear equation (e.g. Campana and Hurley, 1989; Suthers *et al.*, 1989; Meekan and Fortier, 1996). However, cod larvae typically grow at variable rates, and otolith and somatic growth may become disassociated (e.g. Campana and Hurley, 1989). Thus, departure from the linear otolith size–fish size relationship is possibly due to the change in growth rate with increasing fish size at metamorphosis, demonstrated for cod (Otterlei *et al.*, 1999). Nevertheless, incorrect assumptions about the model form may cause errors in the back-calculation of fish length from otolith size and prediction of growth rates. A possible means to overcome this problem could be the use of a common intercept method (Campana, 1990; Suthers and Sundby, 1996). As a result of the ontogenetic shift characterized by the onset of metamorphosis, different biological intercepts should be used, however, depending on whether back-calculation of fish size from otolith size is carried out for larval or juvenile cod. The effect of allometry between otolith growth and somatic growth associated with life history transitions like the onset of metamorphosis has previously been demonstrated for some species of flatfish (e.g. Jenkins, 1987), although not for others, e.g. starry flounder (*Platichthys stellatus*) (Campana, 1984).

The otolith radius–fish length relationship was temperature- and stock-dependent. The radius of the lapillus at length significantly increased with increasing
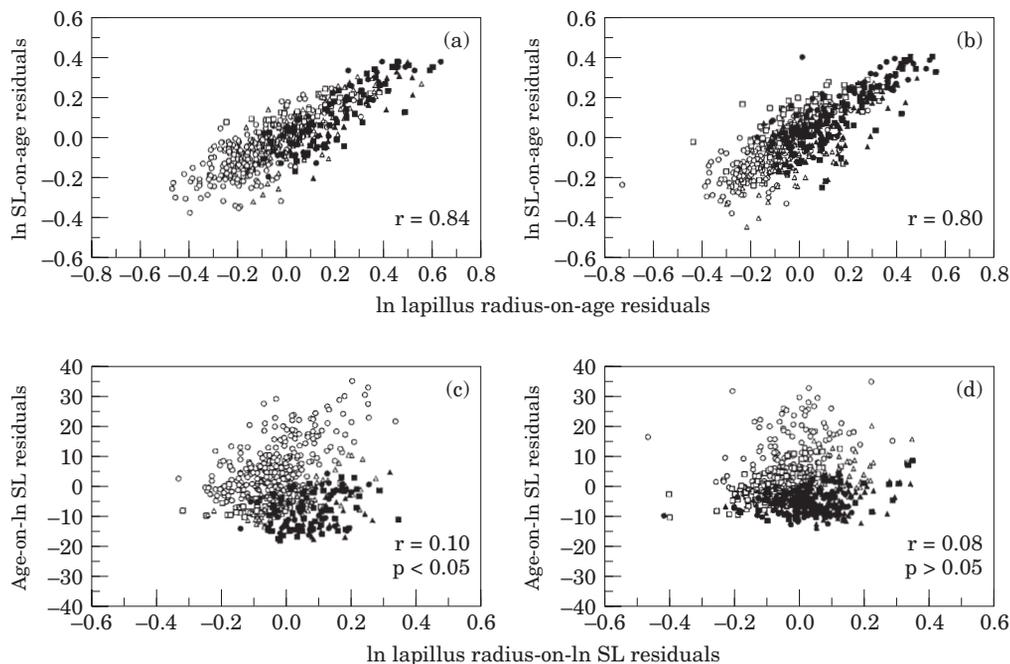
Figure 6. Age-independent variability and growth rate effect, evaluated by correlation analysis of respectively standard length-on-age versus lapillus radius-on-age residuals and age-on-length versus radius of lapillus-on-length residuals for (a and c) Norwegian coastal cod (NC) and (b and d) northeast Arctic cod (NA) larvae from multiple temperature groups. Residuals are generated from a second order polynomial fitted to the data of the different variables. Symbols represent 4°C (○), 6°C (□), 8°C (△), 10°C (●), 12°C (■) and 14°C (▲).

temperature, within both the larval and juvenile stages. The development of larger otoliths by fish raised at high temperature compared to individuals of comparable length raised at low temperature has been reported for several species (e.g. Folkvord et al., 1997; Hoff and Fuiman, 1993). Back-calculated fish sizes can therefore be biased if common relationships are unjustifiably used on fish stocks originating from geographic areas exposed to different temperature regimes, and regional differences in the fish length–otolith size relations should be tested with ANCOVAs before any common relation is used (e.g. Suthers and Sundby, 1993). Moreover, the otolith radius of larval and juvenile NC stock was 5.7% and 4.5% larger, respectively, at given lengths, than the NA stock. Genetic differences among cod stocks are well documented (e.g. Møller, 1968) and differences in otolith macrostructure characteristics of these stocks have been reported (Rollefsen, 1933). On the other hand, no significant differences in otolith growth pattern were found among co-reared cod larvae from different stocks, although cod larvae of northeast Arctic cod remained larger than larvae from Norwegian coastal cod due to their larger initial size (Suthers et al., 1999). In the field however, the different spatial distribution of offspring from different stocks, and their different environmental histories, may warrant the use of the otolith microstructure as a verification tool of different stock units.

Maximum otolith radius of two-day-old larvae was larger than the radius of the hatch check of lapillus, in accordance with the existence of a true hatch check in cod, suggested by Dale (1984). Estimated mean growth of the otolith radius varied from 4.2–4.6 μm d^{-1} at 14°C, but was less than 1.0 (0.4–0.7) μm d^{-1} in both stocks at 4 and 6°C during the first two to three weeks of the experiment. The result implies that the initial average daily otolith increment widths of well fed cod exposed to low temperatures in the laboratory, at common ambient temperatures for these stocks in the field, is below the practical resolution limit of the light microscope (about 0.8–1.0 μm, Neilson, 1992). Unreasonably high estimated growth rates reported for larval cod exposed to low temperatures (<6°C) in the field (e.g. Anderson and Dalley, 2000), may indicate an underestimation of daily increments (and age). Although otolith increments of laboratory-reared larvae are typically less distinct than those from larvae grown under field (i.e. mesocosm) conditions (e.g. Bergstad, 1984), the otolith growth observed in this study suggests that problems with underestimation of increments is very likely at temperatures less than 6°C.

A dome-shaped relationship between mean daily growth of the otolith radius and body size is in accordance with the response described for weight-specific growth rate versus fish size in the present experiments
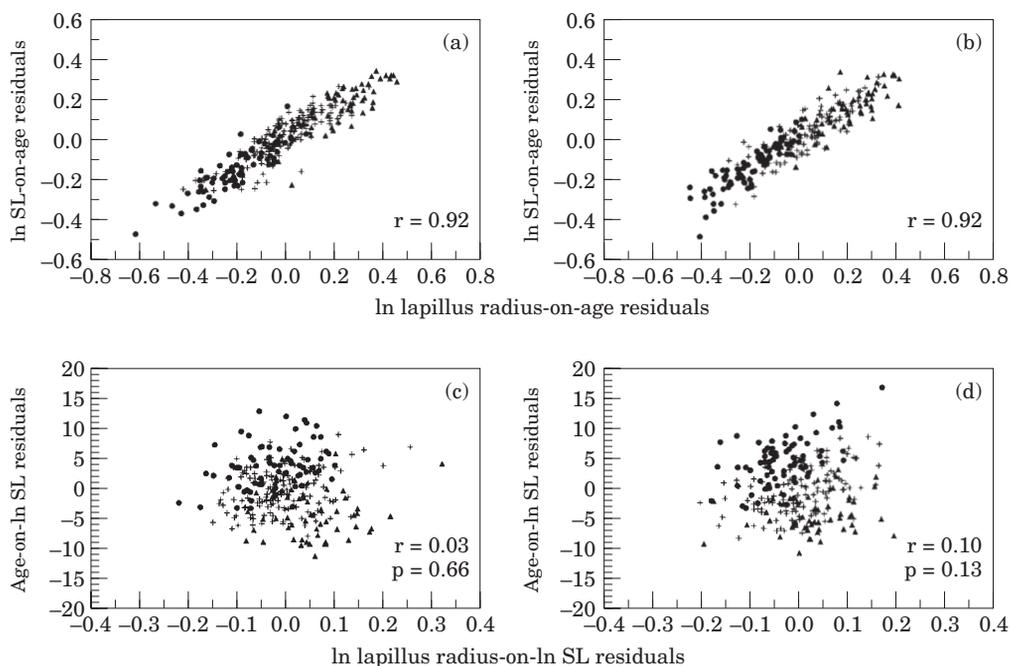
Figure 7. Age-independent variability and growth rate effect, evaluated by correlation analysis of respectively standard length-on-age versus lapillus radius-on-age residuals and age-on-length versus radius of lapillus-on-length residuals for (a and c) Norwegian coastal cod (NC) and (b and d) northeast Arctic cod (NA) juveniles from multiple temperature groups. Residuals are generated from a second order polynomial fitted to the data of the different variables. Symbols represent 10°C (●), 12°C (+) and 14°C (▲).

(Otterlei *et al.*, 1999). However, a peak in the growth rate of lapillus, registered at a fish size of about 25 mm, differs from the size where the somatic growth is maximized, at around 10–12 mm (Otterlei *et al.*, 1999). This may reflect a response lag of otolith growth compared with somatic growth (i.e. the otolith increment width does not always reflect the length or weight increment on a daily basis), observed for herring (e.g. Folkvord *et al.*, 1997) and other fish species (Secor and Dean, 1989).

Finally, this study has demonstrated that the age-independent variability and the growth effect are more distinct when tested temperature-specifically. Effects of growth rate and age-independent variability on the otolith size–fish size relationship have been demonstrated in bluefish (*Pomatomus saltatrix*), and the implications for back-calculation have previously been described in detail by Hare and Cowen (1995). Definitely, the relationship between somatic and otolith growth is complex, influenced by temperature, stock, ontogenetic and growth effects, among others. Current back-calculation techniques should therefore be used with caution and modified to account for these effects.

Acknowledgements

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