THE EFFECTS OF EGG SIZE AND INCUBATION TEMPERATURE ON THE HATCHING AND EARLY GROWTH OF LARVAL HERRING

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Introduction

Egg size variation has been well studied in N.E. Atlantic herring, *Clupea harengus*, stocks (Almatar and Bailey, 1989, Hempel and Blaxter, 1967 and Blaxter and Hempel, 1963). Egg size not only varies between herring stocks which spawn in different seasons and at different geographical locations but also amongst individuals within the same spawning stock. This variation in reproductive strategy between investing more yolk in fewer eggs and investing less yolk in more eggs will affect the characteristics of hatching larvae and therefore their probability of survival. This study investigated various hatching characteristics of three N.E. Atlantic herring stocks.

Several studies have shown that longer larvae hatch from larger eggs (Marteinsdottir and Able, 1992, mumichog; Blaxter and Hempel, 1963, herring). Swimming speed is positively correlated to larval length (Batty and Blaxter, 1992) and therefore a larvas ability to feed and escape from predators will be affected by initial egg size. Larvae have also been shown to hatch with larger yolk reserves and more body tissue from larger eggs (Beacham and Murray, 1985, Chum salmon; Blaxter and Hempel, 1963, herring). Increased body reserves would be expected to increase the time to starvation of a larva (Blaxter and Hempel, 1963) and give the larva a better chance to encounter a patch of food at suitable density before total yolk sac resorption. However, Chambers *et al* (1989) found no relationship between the quantity of yolk reserves of capelin and the expected post hatch survival time in the absence of food.

Temperature on herring spawning grounds varies considerably from year to year. For example a 42 year series of temperatures taken from the North Channel, an area close to Ballantrae Bank (the spawning site of the Clyde herring used in this study), shows that the mean temperature during March ranged from 4.8-9.8°C (Jones and Jeffs, 1991). The temperature that eggs and larvae experience will affect development by altering the rates of many biochemical and physiological processes (Blaxter, 1993).

Temperature can act directly to effect the survival of embryos even within their zone of tolerance, both higher survival at lower temperatures (Forrester and Alderdice, 1966, Pacific cod) and an optimal survival temperature (Beacham and Murray, 1985, Chum salmon) have been reported. Temperature also has a direct effect on time to hatch (Hempel and Blaxter, 1967). Egg size has been reported to both interact with temperature to affect hatch time (Pauly and Pullen, 1988) and have no effect on the temperature hatch time relationship (Miranda et al, 1990, sardine; Beacham and Murray, 1985, chum salmon; Blaxter and Hempel, 1963, herring).

Incubation temperature is reported to have a range of effects on the hatch length of fish larvae. Embryos are either not affected by incubation temperatures (Blaxter, 1956, Clyde herring) longer at hatch when incubated at a higher temperature (Forrester and Alderdice, 1966, Pacific cod; Blaxter and Hempel ,1963, Clyde herring; Blaxter, 1956, Buchan herring), or longer at hatch when reared at a lower incubation temperature (Beacham and Murray, 1985, Chum salmon; Blaxter and Hempel, 1961, German coastal herring and Meyer, 1878, Baltic herring).

The above studies largely look separately at the effect of either temperature or egg size on the developing embryo. This study aims to investigate the effect of the interaction between egg size and temperature on: survival; time to hatch and length, weight and yolk volume on hatch.

Materials and Methods

Percentage survival

Ripe adult herring were caught off their spawning grounds and embryos incubated in the laboratory following the techniques of Blaxter (1968). 10 eggs were counted from each female, dry weights measured and egg batches covering a wide range of egg sizes were selected for each stock. Larvae were incubated at the temperatures shown in table 1.

Stock	Year	Temperature regimes/°C	Egg dry weights, mg
Buchan (Bu94)	Autumn 1994	8, 12 and 15	0.12-0.19
Manx (Mn94)	Autumn 1994	10 and 13.5,17	0.28-0.4
Clyde (Cl94)	Spring 1994	5, 8 and 12	0.28-0.39
Clyde (Cl95)	Spring 1995	5, 8 and 12	0.3-0.44

Table 1. Incubation details

Three replicate microscope slides of Buchan eggs for each of 32 females and at each temperature were photographed at intervals from fertilization until hatching. In each photograph the number off eggs surviving as a proportion of the number of eggs fertilized was counted.

Hatching characteristics

Before hatching eggs were transferred to 1 litre floating cylindrical containers with a 63μm mesh floor to facilitate water exchange. Larvae were removed daily, counted and a sub-sample of, where possible, 10 larvae from each egg batch were measured for the following parameters: total length; the maximum yolk sac width and length (from which yolk volume was calculated using equation 1); and dry weight.

Equation 1:

$$\frac{4}{3}\pi * \frac{1}{2}(yolklength) * \frac{1}{2}(yolkwidth)^2$$

Daily measurements were continued until peak hatching and mean values on the day of peak hatch were used for further analysis.

From the data of the number of larvae hatching on each day the date of 50% cumulative larval hatch was calculated and this was rounded up to the nearest day. Greater accuracy could not be used as larvae could only be counted daily as they hatch at night. The SAS procedure GLM (SAS Institute Inc., 1988) with an analysis of covariance model was used to test the significant factors affecting the hatch date distribution, and each of the three hatching characteristics, total length, dry weight and yolk volume.

Variable name	Variable type	P>F
1/temp.	continuous	<0.01
Stock	class	0.20
Egg size	continuous	0.70
Overall model		<0.01

Table 2. The results of a SAS GLM analysis of covariance model on the date of 50% hatch of larvae. No interaction terms were significant.

Results

There was no clear relationship between egg dry weight and percentage survival of Bu94 eggs incubated at 15, 12 or 8°C. Survival of eggs reared at 8°C, 25.12±12.04 (±1S.D) was generally higher than that of fish reared at either 12°C, 6.71±7.40, or 15°C, 2.73±3.45.

The SAS GLM model showed that there were no effects of stock or egg size on the date of 50% hatch of larvae, Table 2. Time to hatch, for all stocks, was related to the inverse of incubation temperature, Equation 2.

Equation 2:

$$HatchTime = \frac{140.01}{incubationtemperature} - 0.71$$

Stock	Variable name	Length	Weight	Yolk volume
Bu94	Temp.	<0.01	<0.01	<0.01
	Egg size	<0.01	<0.01	<0.01
Mn94	Temp.	NS	NS	NS
	Egg size	<0.01	<0.01	<0.01
Cl94	Temp.	<0.01	NS	0.02
	Egg size	<0.01	<0.01	<0.01
Cl95	Temp.	<0.01	<0.01	0.05
	Egg size	<0.01	<0.01	<0.01

Table 3. Results of SAS GLM procedure for each stock separately. Temp. = incubation temperature (a class variable) and Egg size is a continuous variable. NS = non significant variable. All models were significant to P<0.01.

In all cases longer larvae hatched from larger eggs, Figure 3 and Table 3, and the length of larvae at peak hatch also depended upon the incubation temperature in all cases except Mn94, Figure 3b. For a given egg size, embryos incubated at the high temperature were shorter than larvae incubated at the medium temperature which in turn were shorter than larvae reared at the low temperature.

Figure 3. The effect of egg dry weight and rearing temperature on hatching length for a) Bu94 b) Mn94 c) Cl94 and d) Cl95.

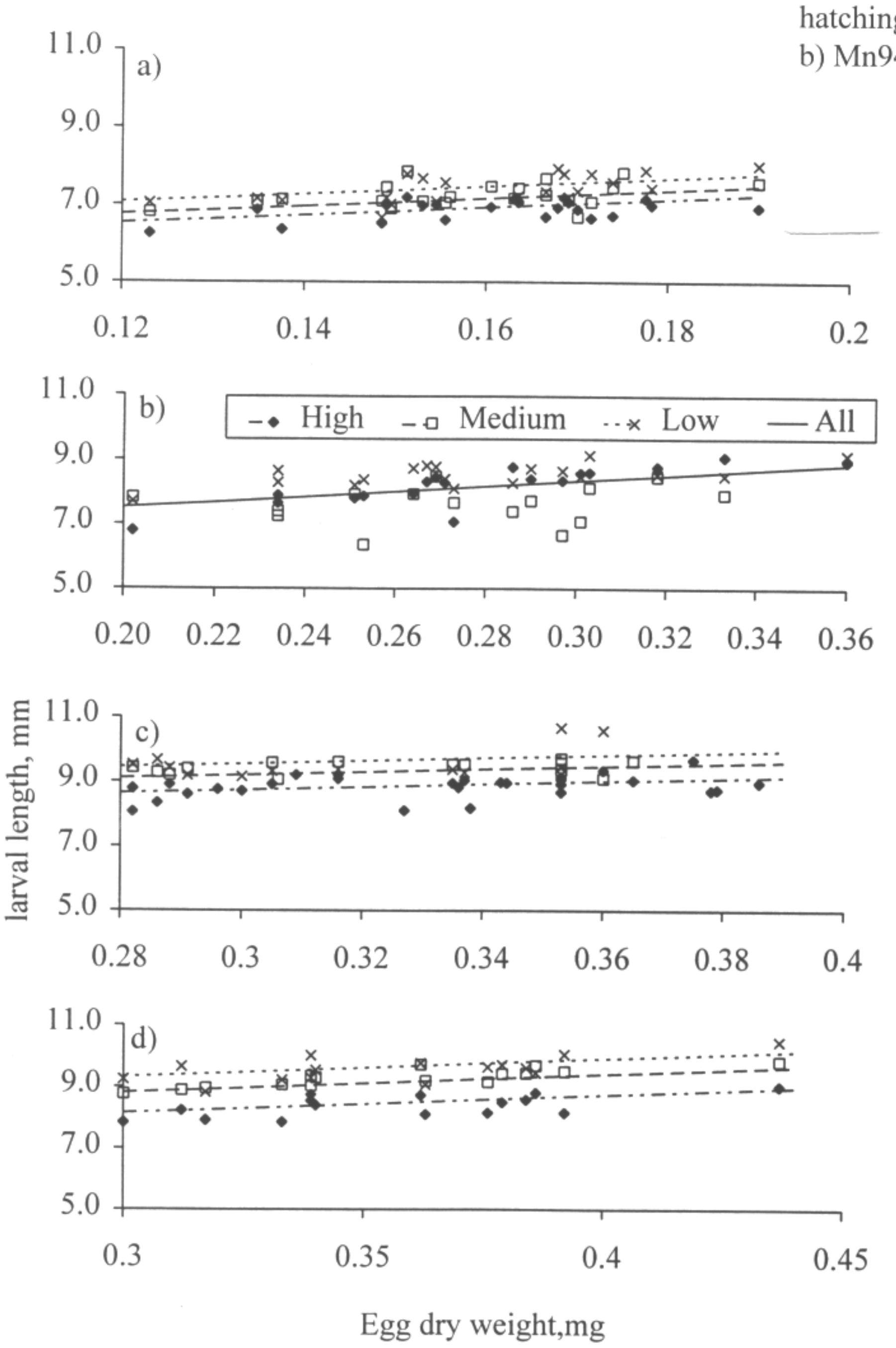

Figure 4 and table 3 show that for all stocks heavier larvae hatched from larger eggs. Also, for a given egg size, Bu94, Figure 4a and Cl95, Figure 4d, larvae were heaviest at hatch when incubated at the low temperature whilst those incubated at the high temperature were the lightest.

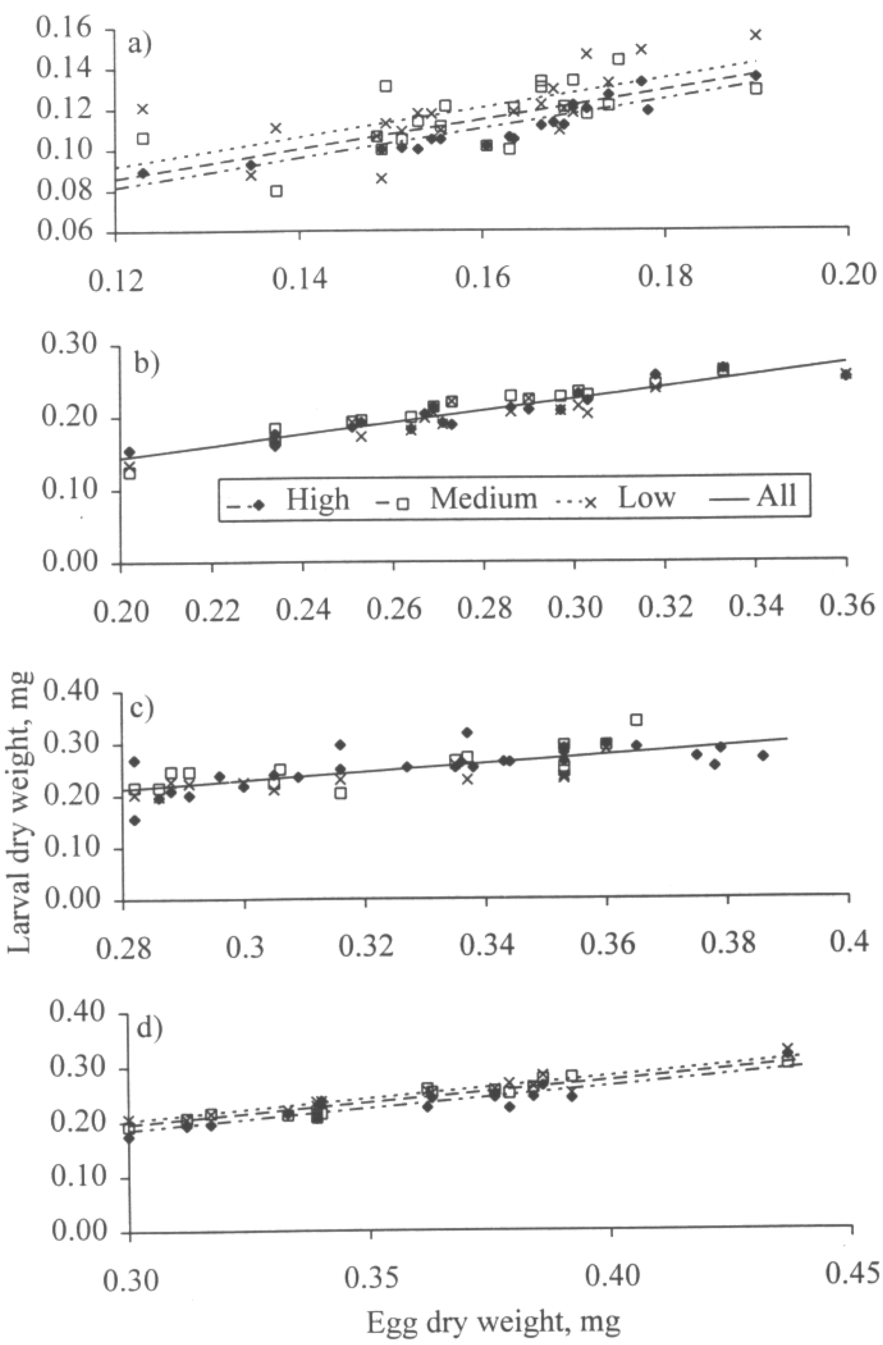

Figure 4. The effect of egg dry weight and incubation temperature on hatching weight for a) Bu94 b) Mn94 c) Cl94 and d) Cl95.

Figure 5 and table 3 show again that in all cases larvae hatched from larger eggs had more yolk. There are again small effects of incubation temperature on the amount of yolk larvae hatch with, but again not for Mn94 embryos. These effects are also reversed between Cl95 and the other two stocks. Bu94 and Cl94 embryos incubated at the low temperature hatched with the most yolk whilst low temperature Cl95 larvae hatched with the least yolk. The same but reverse effect was true for larvae incubated at the high temperature.

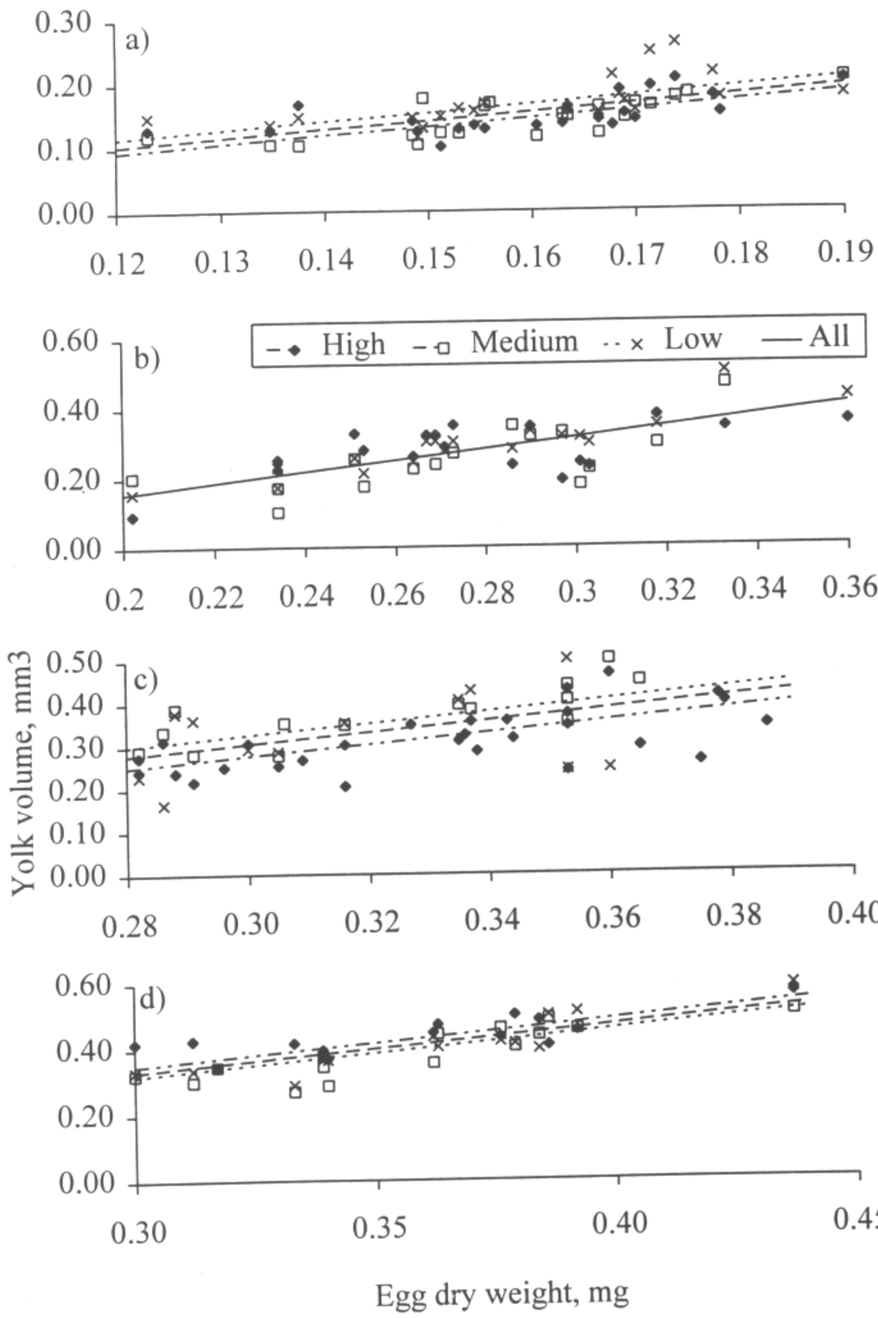

Figure 5. The effect of egg dry weight and incubation temperature on hatching yolk volume for a) Bu94 b) Mn94 c) Cl94 and d) Cl95.

Variable name	Variable type	Ln (length)	Ln (weight)	Ln(yolk volume)
Temp	Class	<0.01	0.20	0.33
Stock	Class	0.84	0.63	0.29
Ln(egg)	Continuos	<0.01	<0.01	<0.01

Table 4. Significance values for SAS GLM analysis of covariance models for the effects of rearing temperature (Temp) stock and egg size (Ln(egg)) on total length, weight and yolk volume.

Table 4 shows that SAS GLM models can be fitted to the ln(length), ln(weight) and ln(yolk volume) data where there is no effect of stock. Ln(egg size) has a significant effect on all three characteristics whilst temperature only had an overall effect on length, Figure 6. The longest larvae hatched when incubated at the low temperature and the shortest larvae hatched from the high incubation temperature, Figure 6a. The fitted line for hatch length has a slope of 0.30 and therefore egg size has a greater effect on hatch length for the smaller eggs in this study. The fitted line to the egg size/hatching dry weight relationship has a slope of 1.00 so an increase in egg dry weight of 1 mg will result in an increase in larval weight of 1 mg, Figure 6b. The fitted line for yolk has a slope of 1.16. Larvae therefore hatch with an increasing proportion of the egg yolk in the form of yolk sac with increasing egg size.

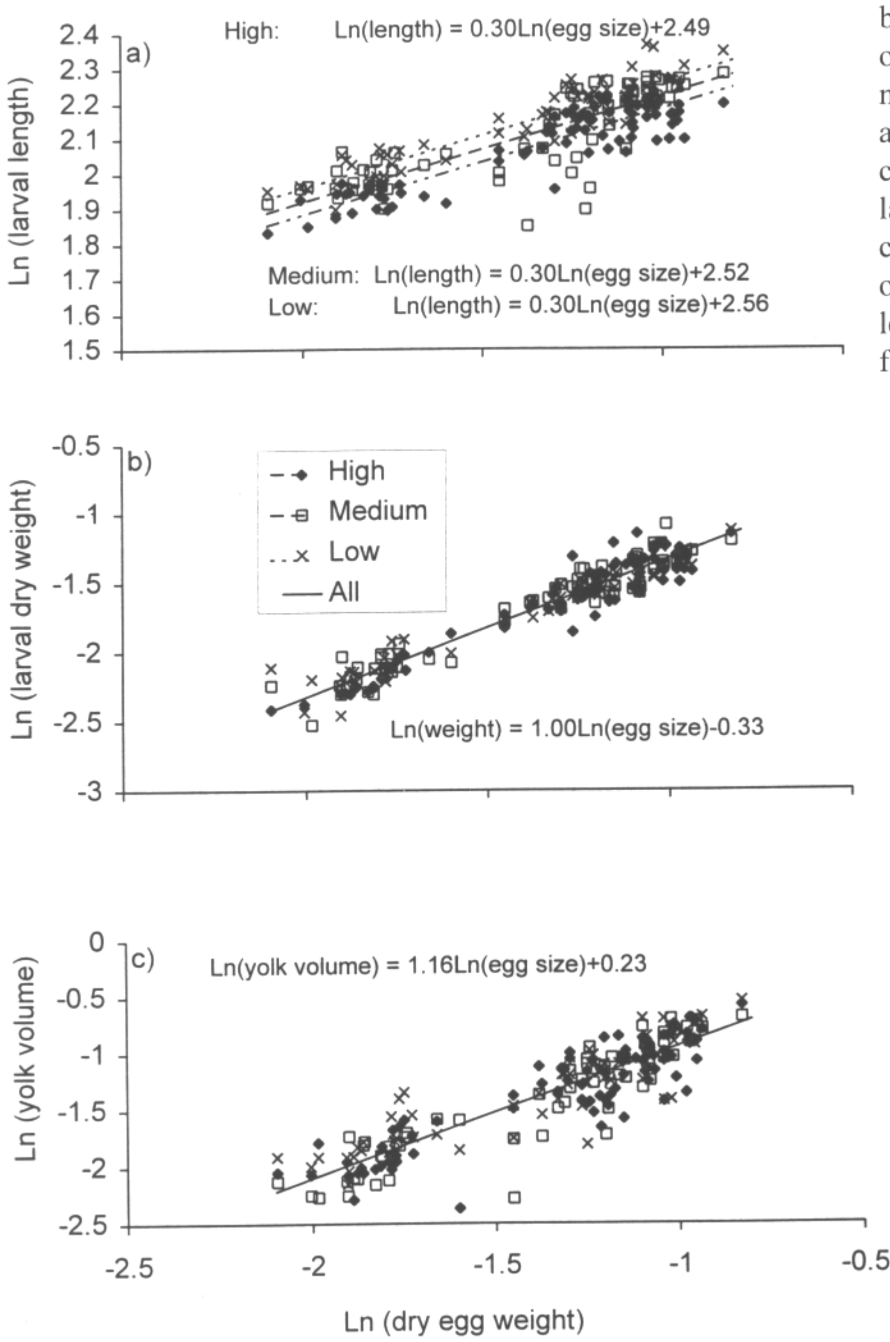

Figure 6. The relationship between natural logarithm of egg dry weight and the natural logarithms of
a) length, b) weight and
c) yolk volume, of hatching larvae from all stocks combined. Temperature only had an overall effect on length. SAS GLM model fits are shown.

Figure 7 shows that for Cl95 larvae which were reared on beyond hatching larvae reared at low temperature generally remain longer for a given weight whilst larvae reared at high temperature remain generally shorter for a given weight.

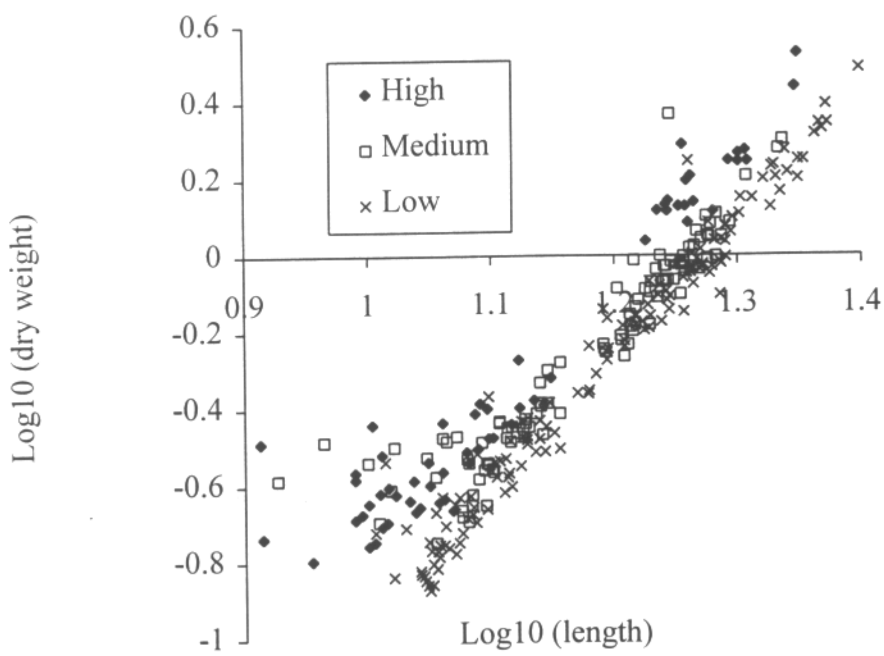

Figure 7. The length weight relationship of larvae incubated and then reared at three temperatures.

Discussion

Incubation temperature had a very similar effect on the timing of hatch to that found by Blaxter and Hempel (1963). Larvae incubated at 14.5°C hatched between 6 and 10 days post fertilization (Blaxter and Hempel recorded a value of 7.5 days at 14°C) larvae incubated at 5°C hatched between 23 and 25 days post fertilization (Blaxter and Hempel recorded a value of 24 days post fertilization).
In addition we have demonstrated that both egg size and spawning stock do not affect the hatching time nor was there any interaction between temperature and egg size affecting the hatching time.

Survival of Manx embryos was highest at the lowest incubation temperature (10°C). Although they suspected that all their treatments were affected by hypoxic conditions Forrester and Alderdice (1966) suggest that survival of pacific cod was higher at lower incubation temperatures because of the lower oxygen requirements of larvae developing more slowly at these temperatures. Frequent water exchange meant that hypoxic conditions were not a factor during our study. If eggs had been incubated at progressively lower incubation temperatures then survival would eventually decline when the temperature drops below the embryos zone of tolerance.

Munk and Rosenthal (1983) showed that egg density had affected the hatching characteristics of Baltic herring. They suggested that the contact area of each egg with the surrounding water will determine the oxygen available to that embryo and markedly affect the embryos physiology and subsequent hatching characteristics. In this experiment eggs were scattered randomly over glass plates to avoid large clumps. However, there could still be some differences in oxygen availabilities between embryos which might account for some of the variation in the relationship between egg size and hatch characteristics in this experiment.

There was a strong relationship between egg size and the three hatching characteristics, total length, weight and yolk volume which could be described by one relationship for all stocks. Length has an asymptotic relationship with egg size which therefore has a steeper slope for smaller egg sizes. Therefore, small changes in egg size have more effect on hatching Buchan larvae than they do on Clyde larvae. This suggests a mechanism where by stocks laying small eggs would have larvae with a wider range of hatching characteristics perhaps making them more likely to produce at least some larvae capable of surviving in widely fluctuating environments. Blaxter and Hempel (1963) found that the Baltic herring stock which have the smallest eggs also have the highest variability in larval size at hatching. The fact that the increased weight of larger eggs is transferred directly into larval weight suggests that whilst in the egg embryos are able to convert the energy stored as yolk quite efficiently. However, this study also found that a greater proportion of the weight of these heavier larvae remains in the form of unutilised yolk still in the yolk sac. This could be an adaptation to give them longer to find food before yolk sac exhaustion in a poor environment.

Temperature variations will clearly interact with egg size to effect the hatching characteristics of larvae, particularly in the case of hatch length. The lack of an overall effect of temperature on weight and yolk volume on hatching suggests that temperatures, over the ranges tested in this experiment, have no effect on the efficiency of yolk conversion. However, the effect of temperature on weight and yolk volume of hatching larvae for combined stocks will be obscured, to a certain extent, by the differences between stocks. In particular the effect of temperature on yolk volume was reversed between years with the same stock. These inter-stock differences and a lack of an overall pattern are not surprising considering the temporal and spatial variation between the spawning stocks.

Larvae are not only longer at hatch for a given size of egg when incubated at lower temperatures but remain longer for a given body weight during the early growth phase at these same temperatures. This suggests itself as a possible mechanism where larvae subjected to lower temperatures remain longer for a given body weight and are therefore able to attain higher swimming speeds to compensate, to some extent, for their reduced swimming speeds at these temperatures.

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