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The Propagation of Cod *Gadus morhua* L.

DEVELOPMENT AND MORTALITY OF COD (*Gadus morhua* L.) EGGS AND LARVAE IN DIFFERENT TEMPERATURES

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ABSTRACT

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This experiment was carried out as a part of a project concerning thermal pollution from a power plant. The paper deals with development and mortality of cod eggs and larvae in different temperatures (6^{o}-18^{o}C). Eggs and larvae were exposed to higher temperatures for parts of the incubation time and the yolk sac period. The development rate increased with increasing temperatures. The mortality increased significantly above 12^{o}C, but was rather low below this temperature. Newly fertilized eggs exposed to higher temperatures seemed to be affected more seriously than eggs exposed to higher temperatures later in the incubation period. Newly hatched cod larvae exposed to high temperatures showed no significant increase in the mortality rate. The yolk sac period decreased with increasing temperature. Short temperature shocks (at about 10^{o}C for 15 min) did not affect the mortality rate of eggs and larvae significantly.

INTRODUCTION

The present investigation was carried out as part of a project on the influence of high temperatures on marine organisms in connection with thermal effluents of sea water from a nuclear power plant. The aim was to investigate the effect of high temperatures on the development and mortality of fish eggs and larvae. The present paper deals with the results on these stages of the cod. Experiments with both long-term high constant temperatures, and short-term shock temperatures have been carried out at different developmental stages.

MATERIAL AND METHODS

The eggs were artificially fertilized in a small jar with a little seawater added at about 6°C, which is the natural spawning temperature. After 2-3 min the excess of milt was washed out and the eggs were transferred into an aquarium with running seawater with a temperature of 6°C. To get homogeneous egg material only one male and one female fish were used.

The eggs were transferred directly from 6°C to the experimental temperatures of 6°, 8°, 10°, 12°, 14°, 16° and 18°C in the following developmental stages: 1aα (blastula stage), 1bβ (gastrula stage), and 3β (late stage with a developed, but heart not beating). Egg development was defined from a scale described by Westernhagen (1970). Newly hatched larvae were also transferred to the same experimental temperatures.

The temperature was automatically regulated in water baths to an accuracy of ±0.1°C. The aquaria, which were held in the water baths, were perspex cylinders with volumes of 4.9 and 8.9 l. The outlet was through a strainer just above the bottom, the flowrate being c. 0.3 l/min (see Fig. 1).

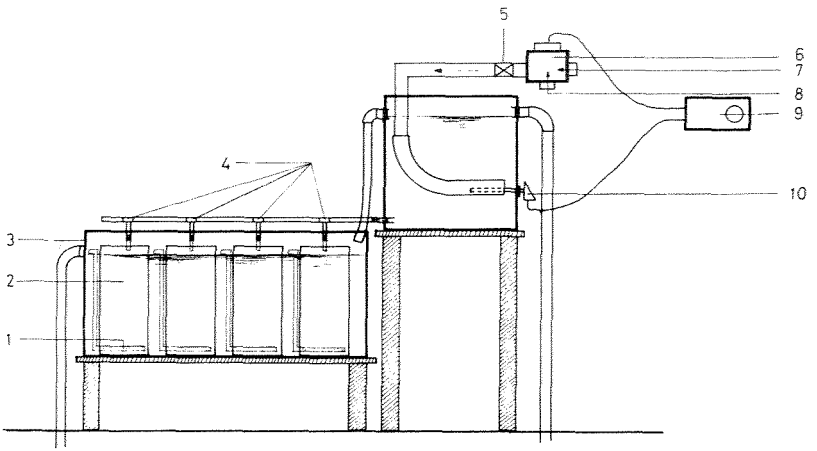

Fig. 1. The experimental aquarium with the temperature regulation unit. 1. Outlet with a filter, 2. Aquarium, 3. Water bath, 4. Water inlet, 5. Tap, 6. Mixing valve, 7. Cold water, 8. Warm water, 9. Regulation unit, 10. Temperature sensor.

The seawater used was pumped in from 75 m depth with a salinity between 35.0 and 35.5°/oo during the experimental period.

The development and mortality were assessed once a day, and dead eggs and larvae removed. The larvae were not fed during the experiment; the experiments therefore continued until 100% mortality, except in 16° and 18°C experiments with eggs from the 3β stage, which had to be stopped earlier due to technical reasons.

In another experimental series, carried out in the same way as mentioned above, the experiments were terminated the day after 100% hatching. The larvae were then examined for abnormalities of body form or behaviour. Dead eggs and larvae were also removed once a day in this experiment and classified as abnormal or not.

To simulate the temperature conditions in the cooling water of a nuclear power plant, eggs in stage 1aα, 1bβ, 3β

and newly hatched larvae were transferred directly from 6°C to aquaria with temperatures about 10 deg C higher. They were held there for about 15 min, and the temperature was then regulated back to 6°C in 15-20 min. Control experiments at 6°C were carried out with eggs and larvae from the same material. The egg experiments were terminated the day after 100% hatching, and the number of abnormal larvae examined.

RESULTS

Egg development

The development of the eggs exposed to the different temperatures from the developmental stages 1aα, 1bβ and 3β are shown in Figs. 2, 3 and 4. Start of hatching (H), 50% and 100% hatched are indicated in the figures. The experiment with the youngest eggs at 6°C was terminated when 50% of the eggs had hatched. In this experiment the developmental rate increased with increasing temperature up to 12°C. At temperatures of 14°, 16° and 18°C no development was observed and the eggs died.

By exposing the eggs to different temperatures from a later, stage 1bβ (Fig. 3), they developed even at 14°C and also to some extent initially at 16° and 18°C. The developmental rate increased with increasing temperature up to 12°C. At 14°C the developmental rate was similar to that at 12°C, indicating a retardation. The eggs exposed to the experimental temperatures even later at stage 3β hatched at all temperatures except 18°C (Fig. 4). Even for the eggs exposed so late, a substantial difference in developmental rate was observed. The time to hatching was about two days at 16°C and five days at 6°C.

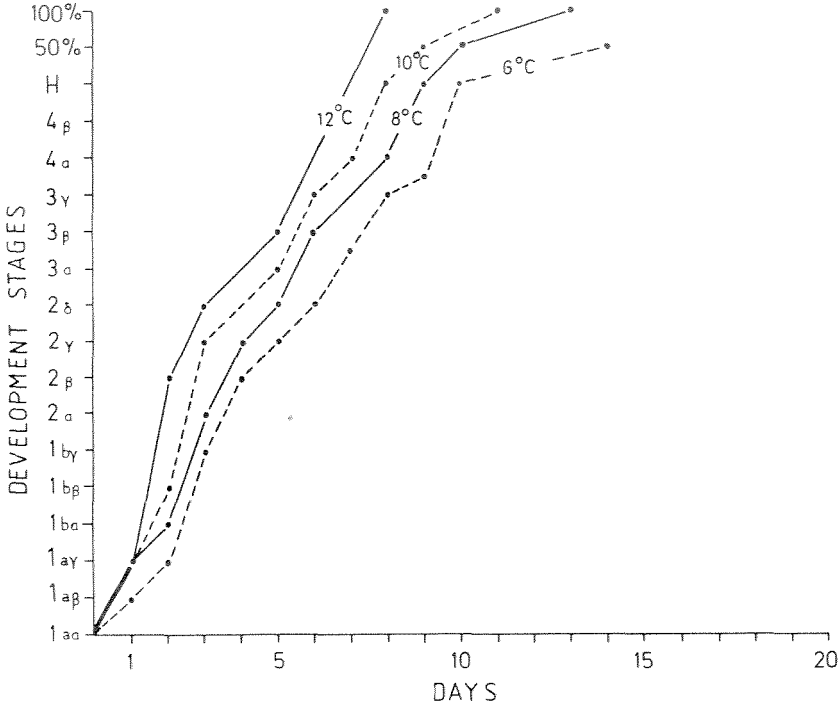

Fig. 2. Egg development rate in different constant temperatures from the first blastula stage (1aα) and onwards. H: hatching begins.

Mortality with temperature

As mentioned earlier, no mortality could be estimated for the eggs exposed at 6°C from stage 1aα. In the same experiments no difference in mortality was observed at 8° and 10°C (Fig. 5). Above this temperature the mortality increased rapidly. At 12°C 80% of the eggs died during the two first days of the experiment, while 100% of the eggs at 14°, 16° and 18°C died within the same period.

Fig. 6 shows the mortality with increasing temperatures for the eggs exposed from stage 1bβ. A higher mortality was

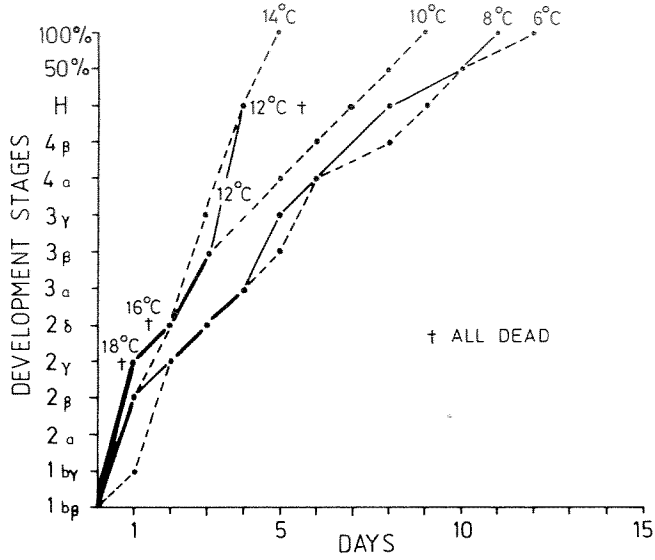

Fig. 3. Egg development rate in different constant temperatures from the gastrula stage (1bβ) and onwards. H: hatching begins.

observed at 8°C than in 6° and 10°C. This was due to an increased mortality between the second and fourth day at 8°C. A similar tendency was observed after the third day at 12° and 14°C, giving a higher total mortality at 12°C. At 16° and 18°C all the eggs died within three and two days respectively. The experiments with eggs exposed from 3β at 14° and 16°C were, as earlier mentioned, terminated before 100% mortality (Fig. 7). The mortality until hatching was similar and negligible for the temperatures 6°-12°C. After hatching had started the mortality increased with increasing temperature due to starvation. At 14° and 16°C the mortality was considerably higher prior to hatching than at the lower temperatures. After hatching the mortality increased rapidly. At 18°C all the eggs died within three days.

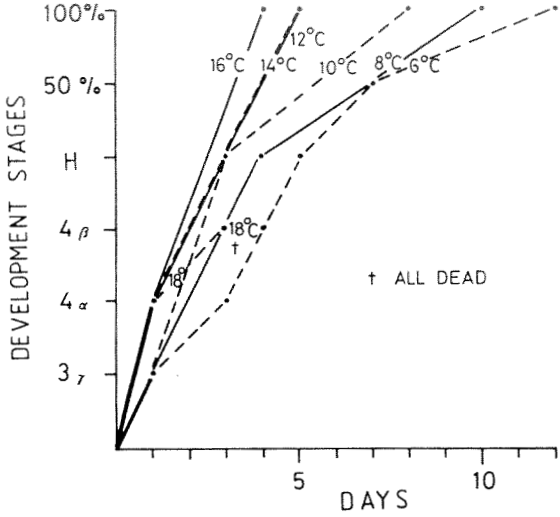

Fig. 4. Egg development rate in different constant temperatures from stage 3β and onwards. H: hatching begins.

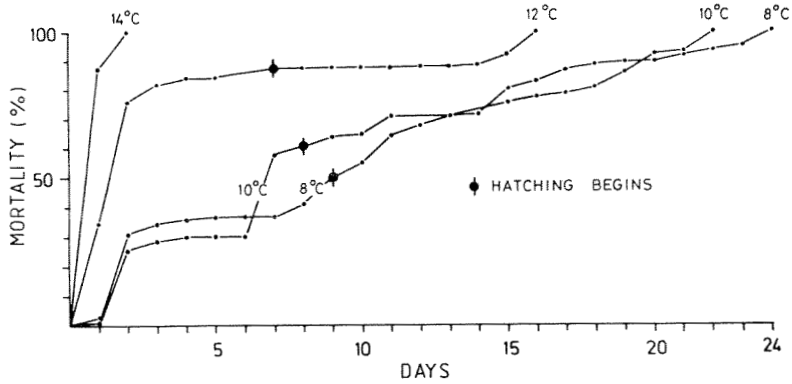

Fig. 5. Egg and larval mortality in the experiments where the eggs were exposed to different constant temperatures from the first blastula stage (1aα).

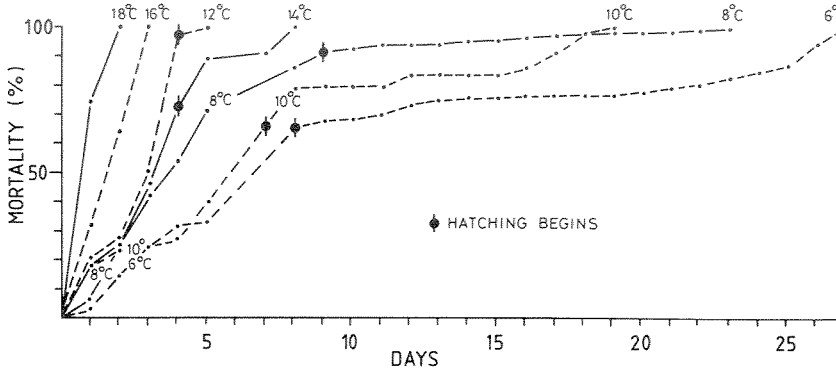

Fig. 6. Egg and larval mortality in the experiment where the eggs were exposed to different constant temperatures from the gastrula stage (1bβ) and onwards.

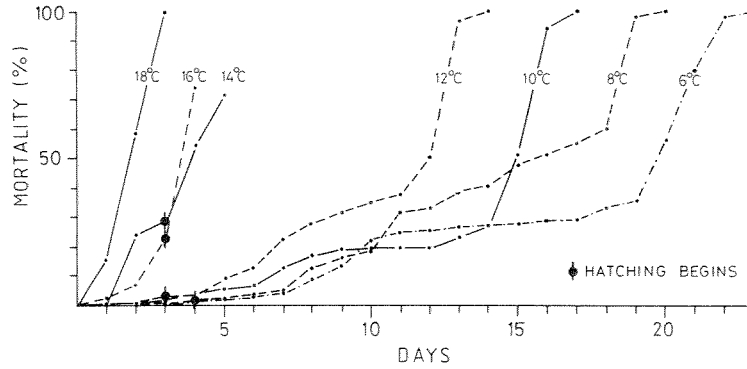

Fig. 7. Egg and larval mortality in the experiments where the eggs were exposed to different constant temperatures from stage 3β.

Mortality in different development stages

Only small differences in mortality in the different development stages were observed at 8° and 10°C for the eggs exposed from stage laα (Fig. 8). At these temperatures the mortality increased after stage laγ, and just before hatch-

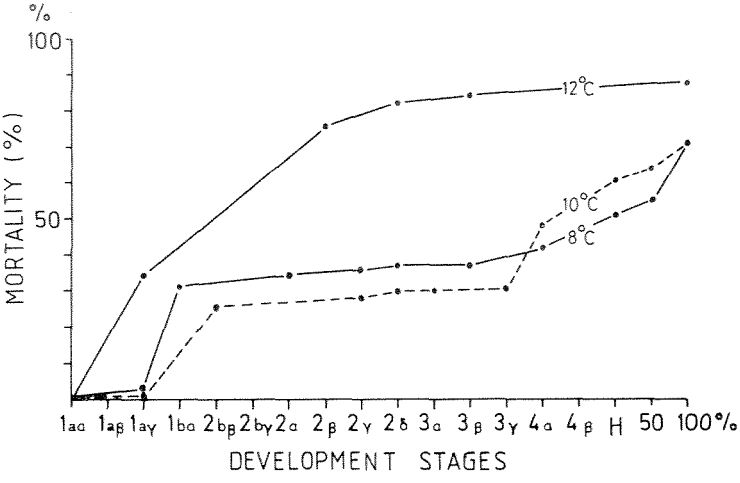

Fig. 8. Mortality in the different development stages for eggs exposed to different constant temperatures from the first blastula stage (1aα).

ing. At 12°C the mortality was high in the stages up to 2β and the mortality was low until the eggs had hatched. All the eggs died without any further development at 14°, 16° and 18°C.

Only small differences were observed at 6°-14°C for the eggs exposed from 1bβ (Fig. 9). The mortality increased steadily at these temperatures until 100% hatching. None of the developmental stages seemed to be more affected by the temperatures 6°-14°C than others. However, at 16° and 18°C 100% mortality was observed during stages 1bβ - 2γ.

Fig. 10 demonstrates the mortality in different developmental stages for eggs exposed to the experimental temperatures from stage 3β. The mortality was low at temperatures 6°-12°C for all stages. At the lowest temperatures the mortality increased slightly during hatching. At 14° and 16°C the mortality was higher and similarly the mortality increased during hatching. At 18°C all the eggs died during stages 3β - 4β.

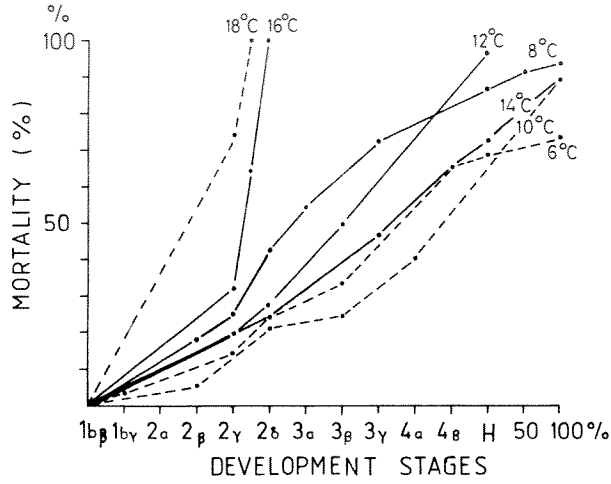

Fig. 9. Mortality in the different development stages for eggs exposed to different constant temperatures from the gastrula stage (1bβ).

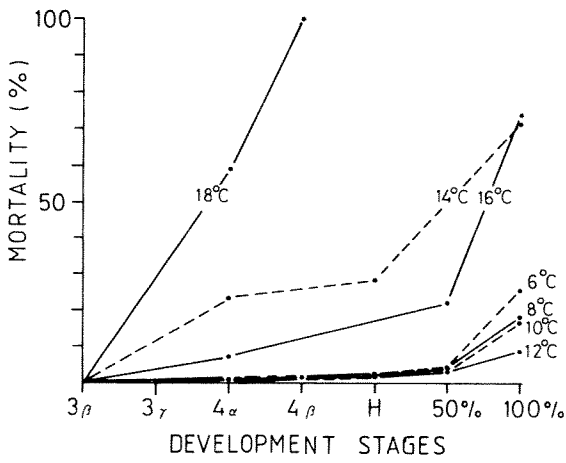

Fig. 10. Mortality in the different development stages for eggs exposed to different constant temperatures from the stage 3β.

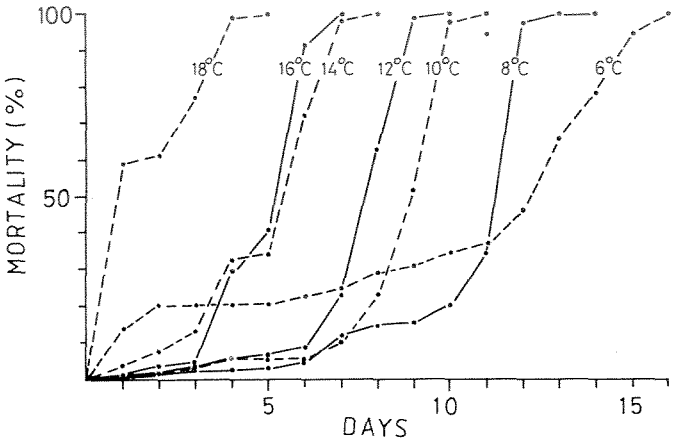

Fig. 11. Mortality of larvae exposed to different constant temperatures from an age of one day.

Larval mortality

The larvae were hatched at 6°C and transferred when one day old to the experimental temperatures. The mortality with time is shown in Fig. 11. At all temperatures, except 18°C, the mortality was rather low until starvation started. At 18°C 60% of the larvae died during the first day and all of them were dead by the fifth day. As demonstrated in Table 1 the resorption rate of the yolk sac increased with increasing temperature. At 6°-10°C the yolk sac seemed to last for the same period, three days. This is probably due to a daily observation interval, which will mask differences less than one day. At 18°C the yolk sac lasted only one day. The period after yolk sac resorption, and until the mortality increased heavily due to starvation, was much shorter at higher temperatures than low ones. At 6°C and 18°C this period lasted for eight and one day respectively. Some of the eggs which hatched resulted in deformed larvae. As shown

TABLE 1

The yolk sac resorption rate and the period of low mortality after yolk sac resorption in the different experimental temperatures.

T°C	Number of days	
	with yolk sac	without yolk sac and with low mortality
6	3	8
8	3	8
10	3	5
12	2	5
14	2	3
16	2	3
18	1	1

in Fig. 12 just a few of the larvae hatched in 6°-10°C were deformed, while the number increased rather drastically at the temperatures above 10°C. A similar trend was observed for the survival rate of the larvae. The survival was much better at temperatures below 12°C than above (Fig. 12). The survival rate and percentage of abnormal larvae were similar whether the eggs were exposed to the experimental temperatures in stages 1bβ or 3β. The percentage of abnormal larvae was also similar for the larvae from the eggs exposed to the different temperatures from development stage 1aα. However, the survival rate was much lower even for the experimental temperatures from later development stages. The survival rate was also much lower in this experiment for the temperatures below 12°C than it was for larvae from eggs exposed to the experimental temperatures at later development stages. This result is difficult to explain but is possibly due to an experimental artifact.

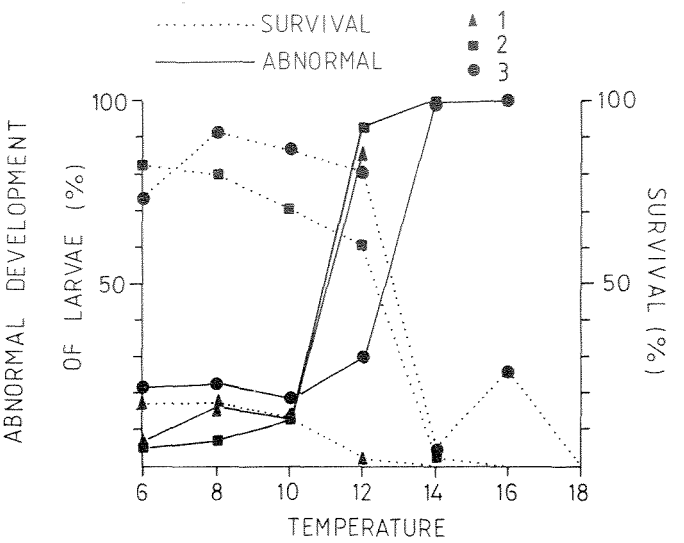

Fig. 12. Percentage of abnormal (solid line) and surviving larvae (dotted line) in different constant temperatures. 1. Larvae from eggs exposed from stage 1aα, 2. Larvae from eggs exposed from stage 1bβ, 3. Larvae from eggs exposed from stage 3β.

Eggs and larvae exposed to temperature shock

Fig. 13 demonstrates that a temperature shock of 15 min with ΔT of 10 deg C had no effect on the eggs in stage 1aα. In this case the mortality in the control group was even higher than in the group given the shock. The eggs which were given the shock in stage 1bβ resulted in an increased mortality of only about 10%. The treatment had no effect on eggs in stage 3β. The percentage of abnormal larvae were low in the three experiments, varying between 6% and 8% in both the shock experiments and the control groups. However, for the larvae the immediate effect was an increased mortality of about 20%.

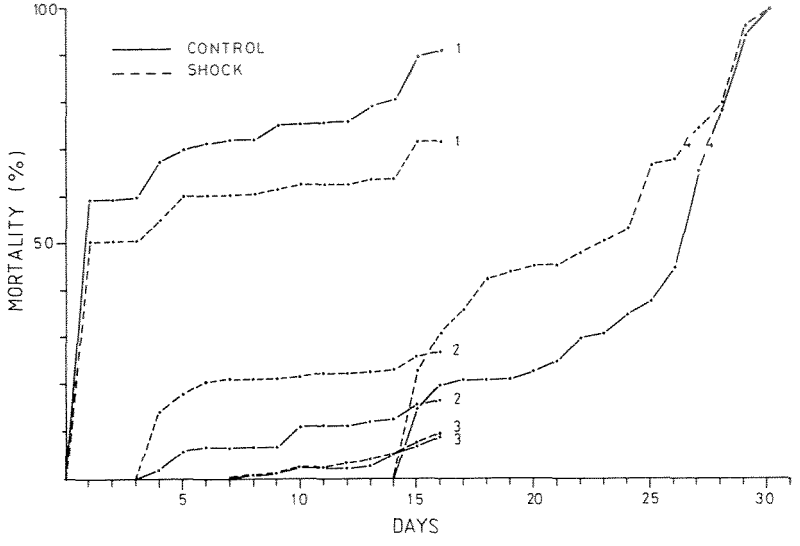

Fig. 13. Mortality in the control (solid line) and the temperature shock experiments (dotted line). 1. Eggs exposed to temperature shock in stage 1aα, 2. Eggs exposed to temperature shock in stage 1bβ, 3. Eggs exposed to temperature shock in stage 3β, 4. Newly hatched larvae exposed to temperature shock.

DISCUSSION

The biological effect of the temperature will vary with varying salinity (Kinne, 1963). Westernhagen (1970) demonstrated that both temperature and salinity could change the mortality rate and the incubation period of plaice, flounder and cod-eggs. According to Holliday and Blaxter (1960) the incubation period of herring eggs was also influenced by the salinity. In the experiments described in this paper the salinity was almost constant, about $35^{o}/oo$ during the experimental period. Therefore the observed effect of the temperature on eggs and larvae is really a combined effect of varying temperature and constant salinity.

The main trend in the different experiments was an increase of developmental rate with increasing temperature. The incubation period of eggs which were exposed to 12°C from stage 1aα and 1bβ was about 35% shorter than observed at 6°C. A similar increase in developmental rate between 6°C and 12°C was also observed for the North Sea cod by Thompson and Riley (1981), and plaice and herring from the Norwegian Skagerrak Coast by Danielssen and Iversen (1974). By increasing the temperature above 12°-14°C the developmental rate did not increase in the same order as increasing the temperature from 6°C to 12°C. A similar effect of retardation in development by increasing the temperature above a certain level was observed for mackerel by Worley (1933) and for cod and plaice by Bonnet (1939) and Westernhagen (1970).

A rather high mortality was observed during the first two days for the eggs exposed from stage 1aα. This was probably caused by a combination of handling, natural mortality and that many eggs were possibly unfertilized. As observed by Laurence and Rogers (1976) no significant differences in mortality rates during the incubation period were observed in the temperature range up to 12°C. In all the experiments the mortality increased drastically for the temperatures above 12°C. However, this increase in mortality was less when the eggs were exposed to the temperatures later in the development. This was also observed for plaice eggs (Danielssen and Iversen, 1974).

The resorption of the larval yolk sac increased with increasing temperature. Only small differences in mortality during the yolk sac stage were observed when the larvae were exposed to temperatures between 6° and 16°C. However, at 18°C the mortality increased drastically. Experiments with newly hatched plaice and herring larvae from the same area (Danielssen and Iversen, l.c.) demonstrated that the yolk sac of the cod larvae was resorbed faster than for these species. A similar tendency was observed for the length of the period after the resorbtion of the yolk sac and until the larvae died due to starvation. The extension of this period was similar for plaice and herring, but shorter for cod. At 6°C

and 18°C the difference was observed to be approximately 10% and 80% respectively.

Abnormal larvae were observed in all the experiments whether the eggs were exposed to the higher temperatures early or late in the development. The number of abnormal larvae was high at temperatures where mortality was high, and low at those temperatures which gave low mortality. The number of abnormal larvae increased considerably at temperatures above 10°C. Laurence and Rogers (1976) concluded that abnormality in cod larvae was independent of temperature up to 12°C. Their experiments were carried out with cod from the east coast of the United States, and that is probably the reason for this difference. They also observed a higher number of abnormal larvae in the temperature range 6-10°C than was observed in the present material. This could be due to both genetic differences and that their experiments were carried out in a closed water system.

The temperature shock of ΔT=10 deg C seemed to have little effect on the eggs and larvae. In two cases, eggs in stage $1a_{\alpha}$ and larvae, the immediate effect was an increase in mortality by about 10% and 20% respectively. However, no effect was observed in the other experiments. In the experiments with 3_{β} eggs the mortality was 10% lower than in the control group. Plaice eggs and larvae treated the same way did not seem to be affected (Iversen and Danielssen, 1977). Similar experiments have been carried out on different fish larvae from the east coast of United States. These experiments demonstrated that a ΔT° of 12 deg C for 40 min had no significant effect (Hoss, Hettler and Coston, 1974). Marcy (1973) investigated some fresh water fish larvae at a nuclear power plant. He observed that 90-100% of the larvae died when they passed through the cooling system and the discharge canal of the power plant. About 80% of the mortality was due to mechanical stress, while the other 20% was attributed to heat shock and retention in water with hign temperature. This is to some extent supported by the present investigation which in two cases demonstrated an increased mortality of 10%

and 20% for eggs in stage laα and larvae exposed to a ΔT=10 deg C.

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