Survival of eggs and yolk-sac larvae of Baltic cod (Gadus morhua L.) at low oxygen levels in different salinities

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In the Baltic Sea, successful spawning of cod is restricted to the deep basins, the Bornholm, Gdańsk, and Gotland basins, with salinities of 10-17. Deep water is exchanged mainly during periods of inflow of saline water from the North Sea. These inflows are irregular and stagnant waters prevail for long periods accompanied by unfavourable oxygen conditions. Consequently, cod eggs occur regularly at low oxygen levels. In this study, batches of eggs (2, 4, and 7 days old) and larvae (newly hatched and 4 days old) were exposed to low oxygen concentrations – 1.0-3.5 mg O₂/l – at two salinities – 11 and 15 – and compared with survival in controls at 100% oxygen saturation in short-term experiments. Egg survival decreased with decreasing oxygen concentration, with no difference in tolerance between egg stages. Egg survival at low oxygen levels was clearly affected by salinity, with significantly higher survival at 15 ppt than at 11 ppt. This implies that egg survival at low oxygen levels differs between the spawning grounds of Baltic cod. It is higher in the Bornholm Basin with higher salinities, 13-17, than in the Gdańsk and Gotland basins, with 10-13. For both larval stages, a marked decrease in tolerance to low oxygen levels was evident between 3.0 and 2.5 mg O₂/l irrespective of salinity, suggesting that the limit for survival of Baltic cod larvae is at about 2.5-3.0 mg O₂/l.


Introduction

Successful spawning of Baltic cod is restricted to the Baltic deep basins – the Bornholm, Gdańsk, and Gotland basins. In the Baltic deep basins there is a halocline at 50–70 m depth with a denser more saline (10–17 ppt) deep water which is only partly mixed with the less saline (6–8 ppt) surface water. The deep water is exchanged mainly during occasions of inflow of saline water from the North Sea. These inflows are irregular, however, and during periods between inflows stagnant conditions prevail with a decrease in the salinity and oxygen content of the water. These varying conditions have a profound impact on the reproductive success of fishes with pelagic eggs. In the Baltic, cod eggs often occur in layers with low oxygen content (see Lebedek, 1978; Wieland, 1989), as determined by their buoyancy.

The year-class strength of Baltic cod is thought to be established during the embryonic period and apparently determined by the thickness of the spawning layer (Grauman, 1973), i.e., the layer with 11 ppt salinity or more (Westin and Nissling, 1991) and sufficient oxygen content for egg and larval development.

The last two major inflows of saline water to the Baltic Sea occurred in the winters 1975–1976 and 1976–1977 (Fonselius, 1988) and resulted in favourable spawning conditions. Consequently, an increase in the Baltic cod stock followed and high catches were reported from the late 1970s until the mid-1980s [450 000 t in the early 1980s (Hansson and Rudstam, 1990)]. However, since stagnant water has prevailed in the deep basins from the late 1970s, there has been a dramatic decrease in the Baltic cod stock, with poor catches reported since the mid-1980s [about 75 000 t reported in 1992 (P.-O. Larson, pers. comm.)].

The metabolic rate of fish embryos is independent of the water oxygen content until a certain critical level is reached, below which metabolism becomes dependent on ambient oxygen concentrations (Rombough, 1986). Any deviation from normal oxygen uptake will influence development and survival negatively (Serigstad, 1987). Metabolic rate in fishes is highly influenced by temperature, since temperature controls the rate of different biochemical reactions, and by salinity, which affects osmoregulation. Consequently, oxygen requirements of eggs and larvae are affected by temperature [see Schur-
The aim of the present investigation was to study the tolerance of Baltic cod eggs and larvae to low oxygen concentrations at prevailing salinities of the Baltic deep basins. In short-term experiments (48 h), eggs (three different stages) and yolk-sac larvae (two stages) were incubated at low oxygen levels; 1.0-2.8 (for eggs) and 1.5-3.5 (for larvae) mg O$_2$/l at 7°C at 11 and 15 ppt and compared with survival in controls at 100% oxygen saturation.

Materials and methods

Spawning cod were caught in gillnets at 50-90 m depth off Gotland (58°N19°E) in the middle of the Baltic Sea. Eggs and semen were obtained by stripping. Fertilization was carried out in water at 7°C and 17 ppt prepared from filtered (0.2 µm cartridge filter) sea water (7 ppt) and synthetic sea salt (hw Marinemix). Eggs at three different stages of development (2, 4, and 7 days old) and larvae at two stages [newly hatched and 4 days old (onset of first feeding)] were incubated. Water of different oxygen concentrations was obtained by bubbling with nitrogen gas to the nearest 0.1 mg O$_2$/l (WTW Oximeter 196). In all incubations, filtered water was treated with antibiotics [Mycostatin (2500 IU/l), Streptomycin (0.05 g/l), and Doctacillin (0.2 g/l)]. Treated water was transferred to the incubation flasks by means of a tube before the eggs and larvae were added. Prior to incubation the eggs and larvae were kept in water at 7°C and 17 ppt salinity.

Batches of eggs and larvae were incubated at five oxygen concentrations, ranging between 1.0 and 2.8 mg O$_2$/l for eggs and 1.5 and 3.5 mg O$_2$/l for larvae, at two different salinities; 11 and 15 at 7°C. As controls, incubations at 100% oxygen saturation (10-11 mg O$_2$/l) in respective salinities were used. Each treatment, containing 125-150 eggs or 10-15 larvae, was incubated for 48 h in a 500 ml flask with conical stopper to prevent air bubbles at the start of incubation. The water was mixed gently three times a day by rotating the flask to prevent microstratification of oxygen. Each set of incubation, i.e., 12 flasks, was performed in replicates with batches from different females as follows: 6 replicates for 2 and 4 days old eggs; 2 replicates for 7 days old eggs; 5 and 6 replicates for newly hatched and 4 days old larvae, respectively. The flasks were placed on their side and the eggs developed at the bottom of 11 ppt, since Baltic cod eggs are non-buoyant at 11 ppt (see Nissling and Westin, 1991); because of positive buoyancy they developed at the top at 15 ppt. At the end of the incubations egg

Figure 1. Survival of Baltic cod eggs incubated at low oxygen levels for 48 h at 7°C with salinities of 11 and 15. Survival in controls is given as absolute survival and survival at low oxygen concentrations is given as relative survival, i.e. as percentage of survival in controls. (a) 2 days old eggs (six replicates), (b) 4 days old eggs (six replicates), (c) 7 days old eggs (two replicates). Standard deviation shown at top of bars.
Table 1. Oxygen concentrations (mg O₂/l) at the start and end of exposure of Baltic cod eggs (2 and 4 days old) (six replicates) and larvae (4 days old) (one larval group) to low oxygen levels. Each experiment was carried out at 7°C with a salinity of 11 for 48 h.

<table>
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<th>Eggs Start of incubation (mg O₂/l)</th>
<th>Eggs End of incubation (mg O₂/l)</th>
<th>Larvae (mg O₂/l)</th>
<th>Larvae End of incubation (mg O₂/l)</th>
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<td></td>
<td>2 days old</td>
<td>4 days old</td>
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<td>10-11 (control)</td>
<td>9.0 ± 0.6</td>
<td>9.3 ± 0.4</td>
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<tr>
<td>2.8</td>
<td>2.6 ± 0.1</td>
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<td>3.5</td>
<td>9.6</td>
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<tr>
<td>2.4</td>
<td>2.3 ± 0.1</td>
<td>2.3 ± 0.1</td>
<td>3.0</td>
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<tr>
<td>2.0</td>
<td>2.0 ± 0.1</td>
<td>1.9 ± 0.1</td>
<td>2.5</td>
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<tr>
<td>1.7</td>
<td>1.6 ± 0.1</td>
<td>1.6 ± 0.1</td>
<td>2.0</td>
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<td>1.4</td>
<td>1.4 ± 0.0</td>
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<td>1.0</td>
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<td>1.1 ± 0.1</td>
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Survival and larval condition were checked. White eggs (dimpled) and eggs which had ceased in development were considered dead and larval condition was judged as follows: dead, moribund (partly white), alive but inactive (did not react when touched), or active (swimming). In order to exclude differences in survival due to differences in egg quality caused by maternal effects and stage during the spawning period (see Kjesbu, 1989; Kjesbu et al., 1991; Solemdal et al., 1991), egg survival was calculated in relation to survival in the controls (survival in the controls treated as 100% survival) and given as a percentage of survival in the controls. To check the consumption of oxygen during the incubation, oxygen concentrations at the end of incubations at 11 ppt were measured.

To examine whether transference from a high salinity (17) to lower salinities (11 and 15) influenced egg survival at low oxygen concentrations, two egg batches were fertilized and incubated at respective salinity until start of exposure, i.e., day 3 (one batch) and day 5 (one batch), and then incubated at low oxygen concentrations at 7°C for 48 h as above.

Results

Table 1 gives the oxygen concentrations at the end of incubations for 2 and 4-day-old eggs and one larval group. The measurements revealed a small decrease in oxygen content during the 48 h incubation. The highest decrease occurred in the controls, while the decrease was low at low oxygen levels. The decreases in oxygen content seem to be related to egg survival, i.e., the higher the survival the higher the oxygen consumption (cp. with Fig. 1).

Egg survival and larval condition after incubation at low oxygen concentrations at 11 and 15 ppt are shown in Figures 1a-c and 2a-d, respectively. Egg survival decreased with decreasing oxygen concentrations at all developmental stages studied, with no apparent difference in tolerance to low oxygen levels between egg stages. Compared with controls (100% oxygen saturation) there was a decrease in egg survival at 2.8 mg O₂/l and only a few eggs survived at 1.0 mg O₂/l. However, egg survival differed between salinities and was significantly higher at 15 ppt than at 11 ppt, p < 0.001 (sign test of paired observations) for both 2 and 4-day-old eggs (the numbers of incubations of 7-day-old eggs were too low for a sign test) at low oxygen concentrations, while no difference in survival occurred between controls at 11 and 15 ppt.

Table 2 gives egg survival at low oxygen concentrations at 11 and 15 ppt for two egg batches incubated at the same salinities from fertilization until the start of exposure to low oxygen concentrations. Egg survival was higher at 15 ppt than at 11 ppt for both 3 and 5-day-old eggs. These results suggest that the difference in egg survival between salinities was not caused by the transference of eggs from a higher salinity (17 ppt) at the start of exposure.

In short-term incubations of larvae at low oxygen concentrations at 11 and 15 ppt, no differences in tolerance to low oxygen concentrations were apparent between controls at 11 and 15 ppt.

Table 2. Survival of Baltic cod eggs (3 and 5 days old) fertilized and incubated with salinities of 11 and 15 and exposed to low oxygen levels for 48 h at 7°C at identical salinities.
Figure 2. Condition of Baltic cod larvae incubated at low oxygen levels for 48 h at 7°C. (a) Newly hatched larvae incubated with a salinity of 11 (five replicates), (b) newly hatched larvae incubated with a salinity of 15 (five replicates), (c) 4 days old larvae incubated with a salinity of 11 (six replicates), (d) 4 days old larvae incubated with a salinity of 15 (six replicates).

Discussion

Surveys in the Baltic deep basins have shown that adult cod may occur in layers with oxygen content as low as 1.4–2.9 mg O₂/l (see Berner and Schemainda, 1957, 1958; Tiews, 1970) and that cod eggs are regularly found at low oxygen levels; 1.7–5.4 mg O₂/l (Lebedek, 1978), 2.6–5.4 mg O₂/l (Wieland, 1989) with high mortalities reported during the egg stage period; 79–98.8% (Grauman, 1973), 99.8% (Wieland, 1987). The critical oxygen

tween larval stages or between incubation salinities. For both newly hatched larvae and larvae at the onset of feeding the majority of larvae were alive and active at 3.5 mg O₂/l, with inactive and moribund larvae beginning to appear at 3.0 mg O₂/l. For both larval stages there was a marked decrease in tolerance to low oxygen concentrations between 3.0 and 2.5 mg O₂/l. Incubations at 3.0 mg O₂/l displayed high survival, whereas incubations at 2.5 mg O₂/l or lower revealed dead or moribund larvae with only a few surviving larvae at 2.5 mg O₂/l.
level for successful development of Baltic cod eggs has been suggested as being 1.4 mg O$_2$/l (Grauman, 1973); >2.9 mg O$_2$/l (Kosior and Netzel, 1989), but experimental data are scarce. (Oxygen levels reported in ml O$_2$/l are recalculated to mg O$_2}$/l).

The present study shows that cod eggs may survive at low oxygen levels for at least two days, but that survival is greatly affected at oxygen concentrations of 1.0–2.8 mg O$_2$/l. Since cod eggs are regularly found at oxygen levels of this magnitude, these results suggest that low oxygen levels in the Baltic deep basins may cause high egg mortalities. As concluded by Wieland, from investigations on cod egg distribution in the Bornholm Basin, vertical occurrence of eggs is not only controlled by egg buoyancy. In addition to water density, the vertical occurrence of eggs is also influenced by oxygen levels as a result of high egg mortality at low oxygen concentrations. Cod eggs were most abundant at depths of 60–75 m at oxygen levels of 2.6–6.9 mg O$_2$/l, but eggs also occurred deeper, at oxygen concentrations of less than 1.4 mg O$_2$/l (Wieland, 1987, 1989).

According to several investigations on marine fish embryos the uptake rate of oxygen increases during development from fertilization to hatching and absorption of the yolk sac [Serigstad, 1987 (cod); Davenport and Lönn, 1980 (cod); Finn et al., 1991 (halibut)], and is also influenced by larval activity level (Davenport and Lönn, 1980; Serigstad, 1987), i.e., an increase of oxygen demand during development. However, in this study no differences in tolerance to low oxygen levels among egg stages or between two larval stages were observed. For both larval stages a marked decrease in tolerance to low oxygen concentrations was evident between 3.0 and 2.5 mg O$_2$/l, with only a few surviving larvae at 2.5 mg O$_2$/l. Accordingly, this suggests that the limit for survival of Baltic cod larvae is at about 2.5–3.0 mg O$_2$/l.

Incubation of eggs and larvae at two different salinities showed that larval condition was not affected by salinity at low oxygen levels, whereas egg survival at low oxygen concentrations was significantly higher at 15 ppt than at 11 ppt. Hence, the higher mortality of eggs at low oxygen levels at 11 ppt compared to 15 ppt, whereas no differences in egg mortality occurred between controls (100% oxygen saturation), suggests that cod eggs are more vulnerable to oxygen deficit at extreme salinities. Further, this investigation indicates that Baltic cod are less affected by extremes in salinity as larvae than at the egg stage.

Although a long-term experiment is required to estimate overall egg mortality from fertilization to hatching at a certain oxygen level, the present investigation indicates a significant difference in total egg mortality between 11 and 15 ppt. For instance, if egg mortality at 48 h of exposure at 2.4 mg O$_2$/l (egg mortality at the three egg stages pooled for 11 and 15 ppt) is extrapolated, it results in an overall mortality (egg development time 13–15 days at 7°C) of about >99.99 and 97% at 11 and 15 ppt, respectively. This indicates a considerable difference in spawning success with salinity for a given oxygen concentration.

The inflows of saline oxygen rich water from the North Sea are irregular and as a consequence conditions for successful spawning vary greatly between years, but also between spawning areas. Because of the relative distance from the North Sea, the most favourable conditions, generally higher oxygen concentrations and more saline water (13–17 ppt compared with 10–13 ppt in the Gdansk and Gotland basins), occur in the Bornholm Basin, which is more regularly influenced by influxes of water from the North Sea than are the Gdansk and Gotland basins, where stagnant periods are more pronounced. Consequently, egg survival is higher in the Bornholm Basin than in the Gdansk and Gotland basins, owing to more favourable oxygen conditions but also, as this study implies, because of higher salinity, since eggs are less vulnerable to oxygen depletion at 15 ppt than at 11 ppt.

Acknowledgments

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References


