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THE RELATION BETWEEN THICKNESS OF CHORION AND SPECIFIC GRAVITY OF EGGS FROM NORWEGIAN AND BALTIC FLATFISH POPULATIONS

By

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ABSTRACT

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Neutral buoyancy, size, and thickness and ultrastructure of the chorion were studied in eggs from populations of flatfish (*Platichthys flesus* and *Limanda limanda*) from western Norway and three localities in the Baltic: Kiel, the Arkona basin and Tvärminne. The observed differences in neutral buoyancy are probably caused by the differing thickness of the chorion and are correlated with the salinity in the area. The differences are thought to be the result of a long term selection process.

INTRODUCTION

Marine teleost species with pelagic eggs are often distributed over a wide range of salinities (APSTEIN 1910, KINDLER 1941). Brackish water populations of such species produce eggs of a lower specific gravity, capable of floating in water of reduced salinity. Usually, but not invariably (see e.g. MIELCK and KUNNE 1932), such eggs are bigger than those of marine populations of the same species.

Most authors have explained the low specific gravity of pelagic eggs in brackish waters as a function of the osmotic conditions, either in the ovary (STRODTMAN 1918, SOLEMDAL 1967), or in the ambient medium (JACOBSEN and JOHANSEN 1908, KANDLER and TAN 1965, HOHENDORF 1968). The immediate osmotic effect on the specific gravity is, however, small, and can only explain part of the observed differences between the eggs of marine and brackish water populations. It has therefore been suggested that the existence of pelagic eggs with low specific gravity in brackish water populations is the result of long term selection (SOLEMDAL 1971).

Interspecific differences in the thickness and ultrastructure of the chorion of pelagic teleost eggs have been described (Götting 1966, HAGSTRÖM and LÖNNING 1968, LÖNNING 1972). As the chorion is the



Fig. 1. Sampling localities.

densest part of the egg, such changes have a marked influence on the specific gravity of the egg. To test the hypothesis that intraspecific variation in the chorion between marine and brackish water populations of a given species may be responsible for the observed differences in specific gravity, eggs from flounders [*Platichthys flesus* (LINNAEUS)] and dabs [*Limanda limanda* (LINNAEUS)] caught at Bergen and at different localities in the Baltic have been investigated with regard to size, neutral buoyancy, and thickness and ultrastructure of the chorion.

MATERIAL AND METHODS

Flounders (*Platichthys flesus*) and dabs (*Limanda limanda*) were caught near Bergen in water of 35 $^{0}/_{00}$ salinity and from Kiel Bay, 22 $^{0}/_{00}$ S. Flounders were also collected from the Arkona basin, 17–18 $^{0}/_{00}$ S, and from the area near Tvärminne Zoological Station, 6 $^{0}/_{00}$ S (Fig. 1).

Diameter and neutral buoyancy were determined on normally developing eggs 24 hours after fertilization, using the methods described by SOLEMDAL (1967).

The fixation and embedding procedure for electron microscopy has been described by Lönning (1972). The material was sectioned on a LKB ultrotome III and examined in a Siemens Elmiskop I.

Ultrastructural studies were mainly carried out on eggs fixed 3—5 days after fertilization, and when nothing else is mentioned the micrographs are taken from this material. From the Bergen area unfertilized eggs and eggs fixed immediately after fertilization were also investigated.

RESULTS

Neutral buoyancy and egg diameter have been measured for eggs of populations of *Platichthys flesus* and *Limanda limanda*, living under different salinity conditions, and the results are given in Table 1. From this it can be seen that, in general, neutral buoyancy decreases with salinity, with the exception of the flounders from the most brackish area, Tvärminne, which have eggs of somewhat higher neutral buoyancy than those from the Arkona basin. In the Arkona basin the buoyancy is only just enough to keep the eggs from sinking, at Tvärminne flounder eggs sink and develop demersally. Similarly, egg diameter in both flounder and dab increases with decreasing salinity. Here again the sole exception are the flounders from the Tvärminne area, which have significantly smaller eggs than those from the Arkona basin.

	Locality	Salinity º/00	Neutral buoyancy º/00 S	Pelagic: P Demersal: D	Egg d	liam. mm	Number of eggs	Individuals
Species					Mean	Range		
Platichthys flesus	Bergen	35	31.8	Р	0.884	0.78—0.94	1 760	14
	Kiel	22		Р	0.959	0.89-1.02	339	2
	Arkona	17—18	16.2	Р	1.185	1.01-1.29	1 470	14
	Tvärminne	6	19.9	D	1.008	0.88—1.11	1 723	19
Limanda limanda	Bergen	35	30.9	Р	0.770	0.750.87	332	7
	Kiel	22	17.5	P	0.928	0.84—1.04	485	7

Table 1. Neutral buoyancy and diameter of flatfish eggs from marine and brackish populations.

In electron microscopic studies the chorion of the fish egg is usually described as consisting of a thin outer layer and a thick, lamellar inner layer. The nomenclature of these layers varies among authors; in this paper the purely descriptive terms «outer layer» and «inner layer» will be used.

Recent studies of the eggs of *Platichthys flesus* and *Limanda limanda* from the Bergen area have shown that the chorion of the egg has about the same thickness in these two species, viz. about 2.5μ ; the ultrastructure of the chorion, on the other hand, shows clear species-specific differences (Lönning 1972).

In eggs of *P. flesus* from Bergen the inner lamellar layer of the chorion consists of 6 lamellae and the distance between the lamellae varies, being largest between the mid-lamellae. Outside the lamellar part a reticular layer is present. The outer layer of the unfertilized egg is rather homogeneous whereas in the 3—5 days old larvae this layer is fenestrated or subdivided into several layers (Fig. 2, see also Lönning 1972, figs. 10, 12).

Eggs of *P. flesus* from Kiel Bay show a somewhat thinner chorion, viz. about 1.8μ . Also in these eggs the number of lamellae is 6, but these lamellae seem to be more equidistant than in the Bergen material. The reticular layer and the outer layer are in these eggs more homogeneous and not so distinct (Fig. 3).

Eggs of *P. flesus* from the Arkona basin have a quite different chorion from those from the Bergen and Kiel area. The diameter is only ca. 1.1 μ and there are but three lamellae, of which the two outer are furthest apart. As in the Kiel material the outer and reticular layer are rather homogeneous, but in the Arkona population these layers are thinner (Fig. 4).

In eggs from Tvärminne, finally, the chorion is nearly as thick as in the Bergen material, viz. 2.3μ . The number of lamellae, however, is only 4. Outside the lamellae a rather distinct, reticular layer is present, followed by the outer layer, which is often somewhat subdivided (Fig. 5). As mentioned above the eggs in this population are demersal.

The chorion of the egg of Limanda limanda from the Bergen area has been described earlier (HAGSTRÖM and LÖNNING 1968, LÖNNING 1972). As already mentioned the diameter is 2.5μ . In this species the lamellar part consists of 9 equidistant lamellae and the outer layer is subdivided into several homogeneous layers of different electron density (cf. Fig. 6, which shows an unfertilized egg).

Eggs of *L. limanda* from Kiel (Fig. 7) have a thinner chorion, viz. 1.6 μ . The number of lamellae is 8 or 9, and also in this population the lamellae are equidistant. The outer layer is less homogeneous than in

the Bergen material, and is somewhat broken up. As the Kiel material consisted of 5 days old larvae, some of these differences, especially in the outer layer, may be due to morphological changes during development (cf. LÖNNING 1972).

A synoptic comparison of the data on the ultrastructure of the chorion in the investigated populations of P. flesus and L. limanda is given in Table 2.

Table 2.	Diameter	of	chorion	and	number	of	lamellae	of	flatfish	eggs	from	marine	and
brackish populations.													

		Che	Number of		
Species	Locality	Thickness µ Mean	Number of lamellae	eggs measured	
Platichthys flesus	Bergen	2.5	6	13	
	Kiel	1.8	6	5	
	Arkona	1.1	3	5	
	Tvärminne	2.3	4	6	
Limanda limanda	Bergen	2.4	9	11	
	Kiel	1.6	8—9	6	

DISCUSSION

The low specific gravity of pelagic fish eggs in brackish areas is of survival value as it will cause the eggs to float in water of lowered salinity.

Short term experiments with flounders which were transferred from high to low salinity showed that the specific gravity of the eggs was little affected (SOLEMDAL 1967). Similarly, Baltic flatfish kept for two years in water of full marine salinity still produced eggs of low specific gravity (SOLEMDAL, in press). The differences in neutral buoyancy in pelagic eggs of marine and brackish water populations of the same species can thus not be explained by water absorption by the eggs due to osmotic conditions in the ovary or the surrounding medium.

Of the different egg components, the chorion is by far the densest. In herring the chorion amounts to 15-30% of the total dry weight (BLAXTER and HEMPEL 1963), in place 19-33% (SOLEMDAL 1970).

Generally the Baltic populations have eggs with a thinner chorion than the marine populations (Table 2). In *Platichthys flesus*, fishes from the Arkona basin have the thinnest chorion, while at Tvärminne, in still more brackish water, the chorion is much thicker and has in fact nearly the same value as in the marine populations. However, at Tvärminne the eggs develop at the bottom, so that a thick, tough chorion is a definite advantage.

Both the thin chorion of the most brackish populations, and the thick chorion of the Tvärminne flounders, may thus well be the result of a long term selection process. This hypothesis is supported by the experiments mentioned above, in which it was shown that in Baltic flatfish kept at high salinity for as much as two years the eggs still were of the low gravity Baltic type. Differences in e.g. feeding conditions, hydrography or spawning season in the Baltic populations can thus be ruled out as a possible explanation for the differences in egg structure.

In both *Platichthys flesus* and *Limanda limanda* there is an inverse correlation between diameter of the egg and its specific gravity (Table 1). Such a correlation does not exist, on the other hand, in *Pleuronectes platessa*. In this species the Baltic populations have the smaller eggs (SOLEMDAL, in press). Egg size per se is thus not likely to be the main cause of the observed differences in specific gravity.

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Fig. 2. Chorion with outer layer (0) and inner layer consisting of a reticular part (R) and 6 lamellae (L). 30 000x. *Platichthys flesus* from the Bergen area.



Fig. 3. P. flesus from Kiel Bay. 30 000x.



Fig. 4. P. flesus from the Arkona basin. 30 000x.



Fig. 5. P. flesus from the Tvärminne area. 30 000x.



Fig. 6. Limanda limanda from the Bergen area. 30 000x.



Fig. 7. L. limanda from Kiel Bay. 30 000x.