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Influence of salinity and cadmium on capsule strength in Pacific herring eggs*

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ABSTRACT: Eggs of Pacific herring *(Clupea pallasi)* were incubated at 5 °C in salinities of 5, 20, and 35 ‰, and in cadmium concentrations of 0.05, 0.1, 1, 5, and 10 ppm (20 ‰ S). Bursting pressures of eggs in the eight groups were measured throughout incubation. In general, bursting pressures rose to a primary maximum after fertilization, declined, rose to a secondary maximum, then declined again toward hatching. Rate of attainment of the primary maximum was related to salinity of the incubation medium. Bursting pressures at the primary and secondary maxima reached final values of about 1300 and 700 g, respectively, in incubation salinities at and above 20 ‰. Corresponding egg volumes were greatest in low salinities (5 ‰) and near minimum values in salinities of 20 ‰ or greater (35 ‰). Cadmium in the incubation medium delayed attainment of primary maximum bursting pressures and primary and secondary maxima were reduced to 200–350 g at Cd concentrations near 1 ppm. Egg volumes also decreased with increased Cd concentration. It appears that Ca^{++}/Cd^{++} ratios, depending on salinity and Cd levels in the incubation medium, influence the properties of both the jelly coat and the capsule of herring eggs. Changes in the properties of these layers could make the eggs more susceptible to mechanical damage, particularly at combinations of higher ($\geqslant$ 1 ppm) Cd concentration and lower ($\leqslant$ 20 ‰) salinities.

INTRODUCTION

During a larger investigation of the influence of salinity and cadmium on the development of Pacific herring *(Clupea pallasi)* eggs, effects of these factors on the strength of the outer covering of the herring egg were examined. The non-specific terms "jelly coat" and "capsule" (zona radiata) are used to refer to the two main layers of the outer covering of the herring egg, because of unresolved problems associated with the origin and nomenclature of the envelope of teleost eggs (Anderson, 1967; Ginzburg, 1968).

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A number of subtle and suggested sublethal responses arising from exposure to cadmium have been noted in marine teleost eggs (Rosenthal & Alderdice, 1976). These include a softening of the capsule during development of the egg of Atlantic herring *Clupea harengus* (Rosenthal & Sperling, 1974; von Westernhagen et al., 1974), and of Baltic flounder, *Pleuronectes flesus* (von Westernhagen & Dethlefsen, 1975). Cadmium was taken up rapidly by eggs incubated in cadmium solutions, although most of it was found in association with the outer covering of the egg rather than with the embryo or yolk sac (Rosenthal & Sperling, 1974). It is assumed that cadmium ions attach to binding sites in the mucopolysaccharide "jelly coat" surrounding the herring egg capsule (Rosenthal & Sperling, 1974). Furthermore, fragility of the envelope of eggs incubated in cadmium solutions suggests that cadmium also may be bound to the protein or polysaccharide (Ginzburg, 1968) components of the capsule itself.

Salinity of the incubation medium also influences the response of teleost eggs to cadmium exposure. Cadmium uptake of Atlantic herring eggs is increased substantially in lower salinities (von Westernhagen et al., 1974). Cadmium uptake in eggs of the Baltic flounder (von Westernhagen & Dethlefsen, 1975) and marine garpike *Belone belone* (von Westernhagen et al., 1975) also is increased in lower salinities, but to a lesser extent. Detrimental effects of exposure to cadmium have been shown to increase in lower salinities for all three species (von Westernhagen et al., 1975). Their data show that more cadmium is taken up by eggs in higher cadmium concentrations, that these amounts approach asymptotic levels, and that these levels are highest in lower salinities. The latter observation suggests that there may be competition between Ca^{++} and Cd^{++} for binding sites in the jelly coat or the capsule of the egg. The lower calcium content of dilute sea water presumably would allow more Cd^{++} to participate in binding or complexing with components of the two egg envelopes at higher equilibrium levels. It is assumed that changes in calcium-cadmium ratios also may have an important influence on hardening (Zotin, 1958) of the egg membrane, a process requiring calcium and inhibited by cadmium (Nakano, 1959).

This paper examines changes in bursting pressure, as a measure of capsule condition, of Pacific herring eggs incubated in cadmium-seawater solutions following fertilization in normal (20 ‰ S) sea water.

MATERIALS AND METHODS

Ripe adult herring were hand-selected for condition and ripeness from a trap net on Saltspring Island, Georgia Strait, British Columbia. They were transported in a 600-l tank, cooled with ice, to the Biological Station, Nanaimo. There they were transferred to a circular tank (8-ft diam, 2½-ft depth). Frozen marine plankton were offered to the fish within 24 hr of the transfer; the food was readily accepted, and it was replaced gradually with fine "starter pellet" salmon feed until the fish were feeding on the pellets alone. Small numbers of herring then were segregated by sex, transferred to 780-l tanks in the laboratory, and used as required. The main

stock of ripe herring provided fertile gametes for over 2 months, until water temperatures reached about 10 °C.

The eggs were applied in rows to 50 x 75-mm microscope slides, previously treated wtih a 1 % solution of Siliclad to facilitate later removal of the adhesive eggs. Eggs were applied to slides singly by touching the genital papilla of the female quickly to the slide surface. The technique provided eggs with one contact plane, that on the glass surface, and avoided possible damage to the jelly coat that could occur in attempting to separate egg adhering to one another. The unfertilized egg samples so prepared were stored (Alderdice & Velsen, 1978) in glass slide-staining carriers in 20 ‰ S sea water at 5 °C. Storage time varied between 30 and 60 min. When samples were complete, all were fertilized simultaneously in a dilution of 0.025 ml herring milt per 100 ml 20 ‰ S sea water at 5 °C. Ten minutes after fertilization the eggs were washed in 20 ‰ S sea water and moved to the test tanks. Test conditions were set up in 40-l tanks at 5 °C, providing (a) 5, 20 and 35 ‰ S sea water without cadmium, and (b) 20 ‰ S sea water with 0.05, 0.1, 1, 5, and 10 ppm Cd. Cadmium solutions were prepared from anhydrous $CdCl_2$ previously dried for 24 hr at 60 °C.

Eggs were removed from the slides with an iris microscalpel, at intervals after fertilization and throughout the incubation period, care being taken not to damage the jelly coat. Bursting pressures were obtained for 10 eggs at each interval, daily until 375 hr after fertilization and every second day thereafter until hatching. Bursting pressure was estimated by positioning each egg between two 1-cm² pieces of microscope slide on the top pan of a direct reading, tared 2000-g balance. A micromanipulator mounted rigidly behind the balance pan positioned a probe on the upper glass plate immediately over the egg. Pressure was applied at rates of approximately 5 g/sec for eggs with bursting pressures up to 100 g, and 50 g/sec for eggs with bursting pressures up to 1000 g. At the bursting point the applied pressure (± approx. 5 %) was recorded.

In some tests a few eggs within the first 24 hr were flaccid and highly elastic. They could be flattened, without bursting, and were not recorded. From 24 hr onward there were a few instances where bursting pressures were greater than 2000 g, the limit of the balance's registration. These values, recorded as 2000 g, differed markedly from usual values and tended to distort computed mean bursting pressures. Calculation of median bursting pressures tended to minimize the effect of the outliers while at the same time stabilizing the variances. For graphic convenience, a logarithmic time scale also has been used to illustrate each bursting pressure series.

Ten egg diameters were also measured at each sampling interval in each of the three salinities and five cadmium concentrations. From these diameters, egg volumes were calculated. Herring eggs tend to be ellipsoidal. Two diameters can be seen in the plane of view under a microscope, one a minor (d_1) and one a major (d_2) diameter. Assuming that the minor diameter not measured (d_3) – that vertical to the plane of view – is equal to d_1, then the volume of an egg

$$V = {}^4/_3 \pi [({}^1/_2 d_1)^2 \times ({}^1/_2 d_2)]$$

Egg volumes were compared for the period between days 4 and 10 (at 5 °C) after

fertilization, a period when egg volumes generally were found to be more constant (Alderdice et al., 1979).

RESULTS

Effect of salinity

Capsule strength. The time course of changes in bursting pressure in the three salinities (Fig. 1) includes a rise from fertilization to an initial maximum, a subsequent reduction until 150–250 hr after fertilization, a period of stability or increase to a lower secondary maximum, and a final decrease just prior to and during hatching. In 20 ‰ S at 5 °C, initial maximum bursting pressures were reached

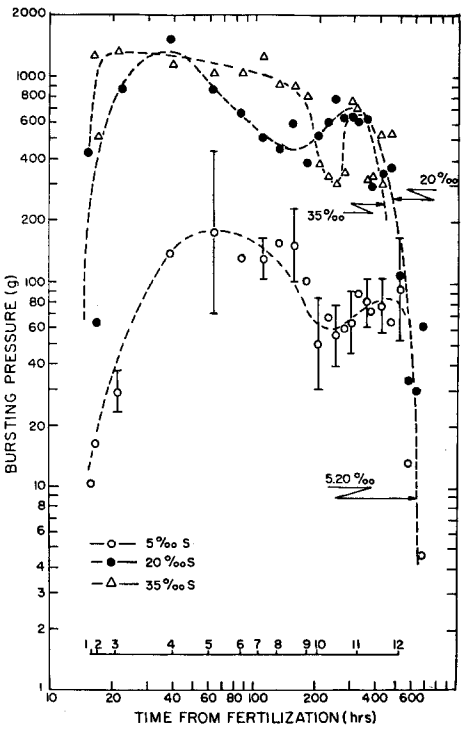

Fig. 1: Trends in change of bursting pressures of Pacific herring eggs fertilized in 20 ‰ S at 5 °C and moved to incubation salinities of 5, 20, and 35 ‰ within 10–12 min after fertilization. Variability in bursting pressures within samples (±1 SD) is shown for the 20 ‰ S trend line. The scale in the lower part of the figure identifies observed stages of differentiation: 1--32-cell; 2--64-cell; 3--early blastodermal cap; 4--late blastodermal cap; 5--gastrula; 6--embryonic shield; 7--blastopore near closure; 8--blastopore closed, Kupffers vesicle prominent; 9--otic capsules evident; 10--embryo encircles yolk, heart beat, embryonic movement; 11--eye pigmentation beginning; 12--embryo two full circles around yolk

in about 40 hr, when embryonic development had reached the late blastodermal cap stage. A following reduction in bursting pressures reached a minimum at approximately 150 hr, after the time of blastopore closure. The secondary, lesser rise in bursting pressure reached a maximum near 300 hr, just prior to the beginning of eye pigmentation and prior to the time when the hatching glands become prominent (360–400 hr). The final decrease in bursting pressure then began, culminating in initial hatches at 620 hr, and maximum hatching frequency near 690–700 hr.

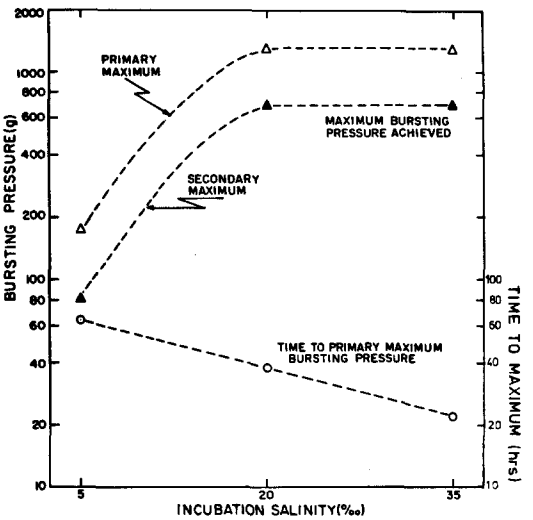

Fig. 2: Bursting pressures estimated for Pacific herring eggs at the primary and secondary bursting pressure maxima, and the time to development of maximum bursting pressures, in relation to incubation salinity

At 5 ‰ S, development of bursting pressure maxima appeared to be delayed in comparison with timing of those events in 20 ‰ S. The initial and secondary bursting pressure maxima occurred at approximately 60 and 450 hr, respectively.

At 35 ‰ S, the initial bursting pressure maximum occurred approximately 20–25 hr after fertilization. The secondary maximum, if such exists, is not well defined. Assuming that the trend at 5 and 20 ‰ also applies at 35 ‰ S, then the secondary maximum would occur at approximately 300 hr.

In general, maximum bursting pressures were about 1300 g, the minimum salinity for full development of bursting pressure being near or somewhat below 20 ‰ (Fig. 2). Bursting pressures achieved in the primary and secondary maxima follow similar courses with respect to incubation salinity. Rate of development of maximum bursting pressure increases with and appears to be a simple function of the incubation salinity.

Egg volume. Average egg volumes between days 4 and 10 of the incubation period were 1.466, 1.384, and 1.381 mm^3 at 5, 20, and 35 ‰ S, respectively.

As might be expected, average egg diameters and volumes were greater in lower salinities.

Related observations. Between 16.0 and 17.5 hr after fertilization (32- to 64-cell stage), the external jelly coat of eggs incubating in 5 ‰ S remained viscous, thick, and adhesive. In 20 ‰ S (64-cell stage) the jelly coat was thinner and harder. In 35 ‰ S (64-cell stage) the jelly coat was thin, hard, hyaline in appearance, and could be peeled away from the egg capsule.

A number of abnormalities were noted in eggs developing in 35 ‰ S sea water. Yolk diameters were noticeably smaller than those in 20 ‰ S at 255–376 hr. Between 325 and 360 hr the embryos were poor in appearance and the finfolds were crenulated. Body length of embryos in 35 ‰ S began to decrease about 424 hr after fertilization, and episodes of embryonic mortality occurred around 279, 424 and 473 hr. In 35 ‰ S, embryonic heart beat was slow and irregular, and body flexures 449 hr after fertilization were limited to rapid, low amplitude movements resembling "shivering" (von Westernhagen et al., 1974). Initial hatching (and maximum hatching frequency) occurred at 570 hr (660–700 hr) (5 ‰), 619 hr (690–700 hr) (20 ‰), and 424 hr (545–570 hr) (35 ‰ S).

Effect of cadmium

Capsule strength. Bursting pressures of eggs fertilized in 20 ‰ S at 5 °C and incubated in 0.05, 0.1, 1, 5, and 10 ppm Cd in 20 ‰ S sea water at 5 °C are shown in Fig. 3. The time course of changes in bursting pressure follows the pattern for the salinity control (Fig. 1, 20 ‰ S) in lower Cd concentrations (0.05, 0.1 ppm), and the trends are modified in higher concentrations (1, 5, 10 ppm). At progressively higher concentrations, cadmium appears to delay development of the primary bursting pressure maximum. The interval lengthened from 30–40 hr after fertilization in the control (Fig. 1, 20 ‰ S) and in 0.05 and 0.1 ppm Cd (Fig. 3), to 40–50 hr in 1 ppm, and to 80–90 hr in 5 ppm. At 10 ppm Cd the primary maximum was eliminated, and bursting pressure continued to rise until the secondary maximum was reached 275–300 hr after fertilization (Fig. 3). The relationship between bursting pressures achieved and cadmium concentration is more complex. Bursting pressures were greatest at the primary maximum for the control and for the 0.05 and 0.1 ppm Cd series. In the remaining higher cadmium concentrations (1, 5, 10 ppm), highest bursting pressures were achieved at the secondary maximum. On the other hand, the magnitude of bursting pressures achieved at both primary and secondary maxima decreased precipitously to levels of 200–350 g with increasing cadmium concentrations to 1 ppm (Fig. 4). The rise in bursting pressure in the 5 ppm Cd series (compared with 1 and 10 ppm) is considered anomalous, an assumption supported by further evidence in relation to egg volume (next section).

Egg volume. Egg volumes were relatively constant between days 4 and 10 following fertilization. During that period measurement of eggs produced the following relation between cadmium concentration and egg volume (mm^3): control (0 ppm Cd), 1.384; 0.05 ppm, 1.386; 0.1 ppm, 1.331; 1 ppm, 1.195; 5 ppm, 1.302;

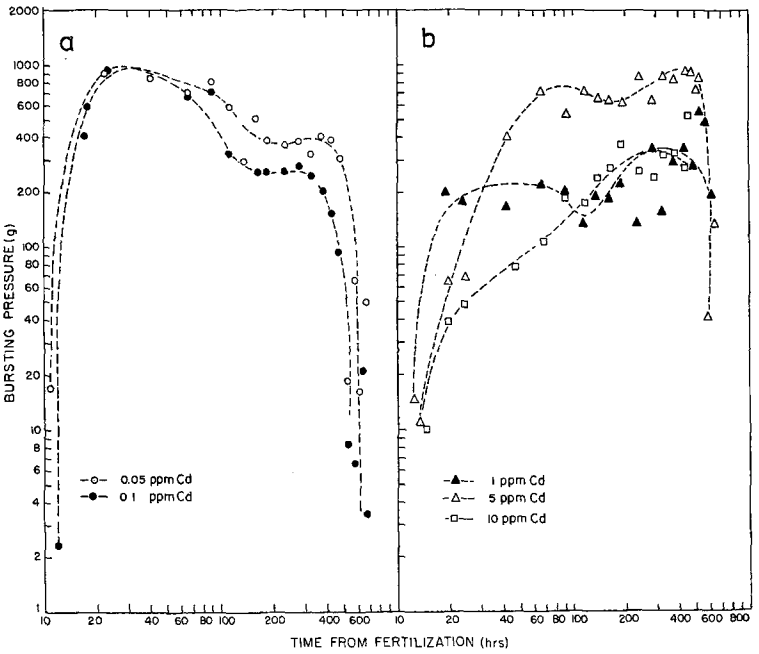

Fig. 3: Trends in change of bursting pressures of Pacific herring eggs fertilized in 20 ‰ S at 5 °C and moved 10–12 min after fertilization to 0.05, 0.1, 1, 5, and 10 ppm Cd in 20 ‰ S at 5 °C for incubation. Scatter in the latter part of the 1 and 10 ppm Cd series is considerable, and the trends are open to interpretation

and 10 ppm, 1.166 mm^3. Again the data for the 5 ppm Cd series seem anomalous. Examination of all the egg volume data for each series from fertilization to hatching suggests that the eggs used in the 5 ppm Cd series were unusually large, leading to the interpretation that in this series both higher egg volumes and higher bursting pressures were atypical. Under that assumption, it appears that increases in cadmium concentration are associated with decreases in maximum bursting pressures and reduced egg volumes.

Related observations. Most ancillary observations were associated with events occurring in the 10 ppm Cd series. In that series, the shape of the blastodermal cap was unusual in comparison with the controls 17 hr after fertilization (Rosenthal, personal communication). The external jelly coat also was less adhesive (33 hr). Epiboly was slightly more advanced (79 hr) compared with that in the other cadmium concentrations, and that slight advance in development was maintained to the stage in which the tail bud of the embryo had lifted away from the yolk sac (200 hr). Embryonic abnormalities were first noted in the head region (127 hr). Embryonic activity decreased substantially between 320 and 345 hr. Shivering movements (von Westernhagen et al., 1974) replaced normal body flexures, as in the series incubated in 35 ‰ S sea water, and swollen pericardia were noted as well as yolksac malformations. In 10 ppm Cd, many embryos appeared to die just prior to hatching. Abnormal embryos also were detected at 378 hr in both the 1 and 5 ppm

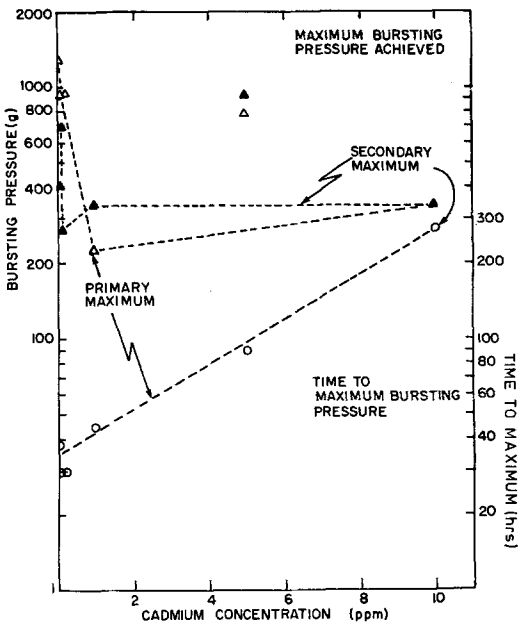

Fig. 4: Bursting pressures of Pacific herring eggs at the primary and secondary bursting pressure maxima, and time required to achieve the maxima, in relation to cadmium concentration at 20 ‰ S and 5 °C. At 10 ppm Cd the primary maximum did not occur; bursting pressures continued to increase with time until the presumptive secondary maximum was reached. Maximum bursting pressures reached at 5 ppm Cd are considered anomalous and have been omitted in the suggested relationship (see text)

Cd test series. A brown discoloration of the jelly coat was observed in eggs exposed to the higher cadmium concentrations (in 1 ppm Cd, after 202 hr; 5 ppm, 60 hr; 10 ppm, 40 hr).

Hatching distributions in the various cadmium concentrations were not obtained. However, the following relation was noted between cadmium concentration and incubation time (hours) to first hatch (and maximum hatching frequency): control, no cadmium, 619 hr (690–700 hr); 0.05 ppm, 618 hr (680–685 hr); 0.1 ppm, 618 hr (680–685 hr); 1 ppm, 523 hr (630 hr); 5 ppm, 476 hr (595 hr); and 10 ppm, 429 hr (450 hr).

DISCUSSION

Development of capsule strength in teleost eggs begins at fertilization. In the flaccid, unfertilized egg the peripheral cytoplasm lies in close proximity to the inner surface of the capsule. At fertilization, cortical alveoli in the cytoplasmic cortex rupture, their contents entering the presumptive perivitelline space (Nakano, 1969). The colloidal alveolar fluid contains organic substances, likely consisting of glycoproteins and lipoproteins (Eddy, 1974). These do not pass through the capsule,

but cause water to be imbibed from the surrounding external medium. The colloids and the imbibed water mix to form the perivitelline fluid. The perivitelline fluid of the Pacific herring egg is detectable about 7 min after fertilization (5 °C); formation of the fluid is half completed in about 2¼ hr and complete in about 6–11 hr depending on the incubation salinity (Alderdice et al., 1979).

Associated with formation of the perivitelline space is a hardening of the egg capsule. Hardening of salmonid eggs is an irreversible process involving changes in properties of the capsule; the process requires calcium (Kusa, 1949) and is attributed to the presence of an enzyme (Zotin, 1958). Even in water-hardened eggs there is a substantial net transport of water across the capsule (Loeffler, 1971). In addition, a number of ions (H^+, Na^+, K^+, Cl^-) pass freely between the perivitelline fluid and the external medium (Shanklin, 1959; Kändler & Tan, 1965 a).

Hence, following fertilization there is an increase in internal pressure as the flaccid egg imbibes water, swells, and become turgid. Loeffler's (1971) evidence of continuous exchange of water between the egg and the external medium implies, by analogy, that the egg resembles an inflated ball with microsieve walls. Internal pressure would be maintained through continuous "pumping" by the osmotic gradient associated with the perivitelline fluid colloid inclusions, which would tend to offset leakage of water and ions through the capsule. By inference from the various studies reported, herring eggs in sea water would set up an internal hydrostatic pressure, influenced by ionic and osmotic gradients, in which the internal turgor pressure would be balanced by the circumferential tension exerted by the elastic properties of the external envelope.

Eddy (1974) summarized these relations for an elastic sphere in equilibrium, whereby the internal pressure (P) of an egg is equal to the product of the tension (T) exerted by the elastic forces of the outer envelope and a function of the radius, $2/r$, so that $P = 2T/r$ (Burton, 1962). Thus a small egg can have a high internal pressure, which can be maintained by a low wall tension. Conversely, a larger egg with the same internal pressure would require a higher wall tension. In the present study, the external force applied to the egg would be transmitted internally to the whole capsule, increasing wall tension. With further increase in pressure, the elastic limit of the egg envelope eventually would be reached and the structure of the envelope would begin to lose its integrity. The bursting pressure is reached when the structure fails and the capsule ruptures. Therefore factors that will influence internal pressure, circumferential tension, or egg volume may influence bursting pressures.

The bursting pressure obtained show similar time trends and primary and secondary maxima as found for eggs of the plaice *(Pleuronectes platessa)* by Pommeranz (1974). Maximum bursting pressures of Pacific herring eggs (1300 g) compare with 400–700 g for the plaice (Pommeranz, 1974; Eddy, 1974), 3000–3500 g for the Atlantic salmon (Hayes, 1949) and 265–450 g for wild carp, *Cyprinus carpio* (Rubtsov, 1973). In general, bursting pressures of teleost eggs examined range from about 100 to 9000 g (Blaxter, 1969; p. 234).

Among marine fishes, egg diameters for a given species tend to be larger in lower salinities (Kändler and Tan, 1965b; Solemdal, 1967; Lönning and Solemdal,

1972; May, 1974). Eggs of the Atlantic herring approach minimum diameters in salinities of 30–50 ‰ (Holliday, 1965). Those of the white sea herring, *C. h. pallasi*, show a continuing decline in diameter in salinities from near 0 to 50 ‰ (Dushkina, 1973). The current results indicate that both egg volume and bursting pressure are influenced by incubation salinity and exposure to cadmium. The question arises as to whether the volume changes noted could account for the changes in bursting pressure observed. Bursting pressure, as measured, estimates the limiting circumferential tension on the egg envelope and that tension is a function of turgor pressure and egg size. It follows that a knowledge of bursting pressure-wall tension relations requires information on turgor pressure. To explore these relations, we incubated another set of eggs in 5, 20 and 35 ‰ S sea water at 5 °C. Turgor pressures were examined with apparatus patterned after that of Gray (1932).

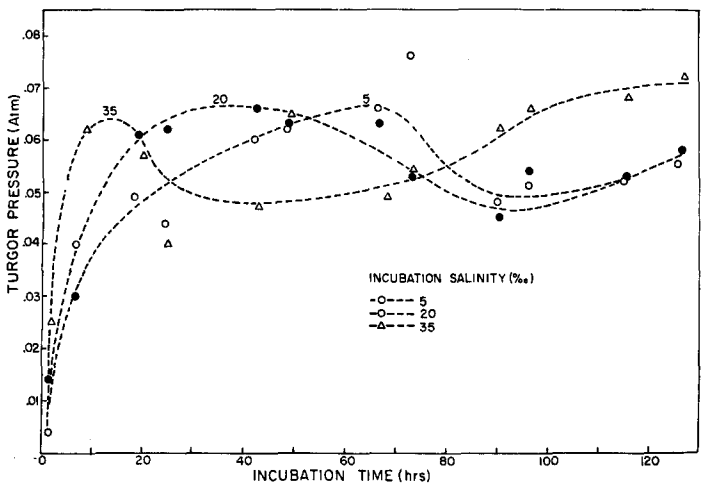

Fig. 5: Turgor pressure (atmospheres) of Pacific herring eggs incubated in 5, 20 and 35 ‰ S sea water at 5 °C. The trend lines are tentative

There is substantial variability in the data obtained (Fig. 5). However initial maximum turgor pressures appear to be achieved more rapidly in the higher salinities in a pattern similar to that for bursting pressure (Fig. 1). Also, turgor pressures of eggs in the three salinities vary approximately between 0.05 and 0.07 atmospheres, the average between 20 and 100 hr being 0.057 atm (43.3 mm Hg), or about 57 750 dynes/cm^2. Two estimates of circumferential tension then may be obtained: (a) that for the egg at rest, based on turgor pressure and size, and (b) the limiting tension, based on bursting pressure of the same eggs. These relations are summarized for the following examples (Table 1).

The anomalous results for the 5 ‰ (0 ppm Cd) and 20 ‰ (10 ppm Cd) incubations, where the limiting tensions are lower than would be expected on the basis of turgor pressure and size of the eggs, suggests that volume changes per se have only a small influence on bursting pressure. Therefore it is presumed that the

Table 1

Influence of salinity and cadmium on egg volume and capsule strength in Pacific herring eggs. Data based on turgor pressure and egg size as well as on bursting pressure

Incubation ‰ (ppm Cd)	Mean egg volume (mm^3)	Radius* (cm)	Surface area (cm^2)	Tension (dynes/cm)	Bursting Pressure (g)	(g/cm^2)	(mm Hg)	Limiting tension (dyn/cm)
5 (0)	1.466	0.0705	0.0624	2035	175	10.9	8.0	396
20 (0)	1.384	0.0691	0.0601	1996	1300	78.1	57.4	2644
35 (0)	1.381	0.0691	0.0600	1995	1300	80.0	57.3	2639
20 (10)	1.166	0.0653	0.0536	1885	350	18.8	13.8	600

* Equivalent spherical radius

influence of low salinity or exposure to cadmium may be expressed through changes occuring in the structure or integrity of the jelly coat or capsule themselves.

Dushkina (1973) noted that the jelly coat ("mucosa") of white sea herring eggs swells greatly in low salinities. Reduction of egg volume in higher salinities was a result partly of thinning of the jelly coat and partly of a decrease in the volume of the perivitelline fluid. Rubtsov (1973) removed the jelly coat from eggs of the wild carp *(Cyprinus carpio)* and compared capsule strength with that of normal eggs with the jelly coat intact. One day after fertilization, capsule strength of eggs without the jelly coat was about one-third lower than in the control. Therefore, the jelly coat appears to contribute to the bursting pressure of the whole egg. In the current study, a delayed thinning of the jelly coat was noted in the low salinity. Hence the delayed rise in bursting pressure and the lower bursting pressure maximum achieved in 5 ‰ S could be attributed in part to a delayed hardening on the jelly coat.

A number of related observations suggest that the structure of either the jelly coat or capsule of marine teleost eggs may be altered by exposure to cadmium. Cadmium inhibits the hardening of the egg after fertilization, a process known to require calcium (Nakano, 1969). Eggs of the Baltic flounder *(Pleuronectes flesus)*, incubated in cadmium-sea water after fertilization in uncontaminated water, developed weakened capsules subject to rupture (von Westernhagen & Dethlefsen, 1975). Similar results were obtained by Rosenthal and Sperling (1974) and von Westernhagen et al. (1974) for eggs of Atlantic herring. In the current study Pacific herring eggs also showed weakened envelopes, particularly in 10 ppm Cd. In general, investigations have shown cadmium to be confined largely to the outer covering of eggs exposed to cadmium (Rosenthal & Sperling, 1974), with levels of contamination being reduced in higher salinities.

The fragility of the outer envelope of cadmium-contaminated eggs draws attention to the need to determine where these effects occur – in the capsule, the jelly coat, or in both. It is assumed that cadmium competes with calcium for binding sites in both the jelly coat and capsule of the egg (von Westernhagen et al., 1975). Cadmium would occupy those sites in relation to Ca^{++}/Cd^{++} ratios, determined by the cadmium concentration, the salinity of the medium, and its pH. The

latter could affect the specificity of the reaction of cadmium with various protein groups because of the different pKs of the latter (Madsen, 1963). Accumulation of cadmium and its toxicity appear related to the presence or absence and thickness of the jelly coat. The thick jelly coat of Atlantic herring eggs accumulates greater amounts of cadmium than the egg of *Pleuronectes flesus* (von Westernhagen & Dethlefsen, 1974), which possesses a much thinner jelly coat. Cadmium also is more toxic to eggs of the latter species, even though the whole egg of *Pleuronectes flesus* accumulates less cadmium than the Atlantic herring egg.

Based on the current evidence, we suggest the following. Eggs possessing a thick jelly coat probably bind in the jelly coat a considerable proportion of the cadmium with which they come in contact. The amount bound likely will be influenced largely by the properties of the incubation medium. The bound cadmium would appear to alter the physical properties of the jelly coat whereby its contribution to the strength of the egg envelope is reduced. In addition, a small amount of cadmium evidently can penetrate the jelly coat and capsule. We suspect that cadmium also must occupy binding sites in the capsule itself, as egg volume is reduced in cadmium-contaminated sea water and there is an apparent loss of elasticity and tensile strength of the capsule.

Finally, the fragility of Pacific herring eggs exposed to very low salinities, or to cadmium-contaminated sea water, could increase the vulnerability of eggs to mechanical damage. For Pacific herring eggs this increase in vulnerability in low salinities would become evident in salinities somewhere below 20 ‰.

ZUSAMMENFASSUNG

Die Druckfestigkeit von Heringseiern erreicht nach der Befruchtung rasch erstes Maximum, fällt dann leicht ab und steigt gegen Ende der Inkubationszeit erneut an. Kurz vor dem Schlupf sinkt die Druckfestigkeit erheblich. Hohe Salzgehalte beschleunigen das Erreichen des ersten Maximums (20–35 ‰ S; 1300 g). In 20 ‰ S tritt es 40 h nach der Befruchtung ein, während es in 35 ‰ S schon nach 20–35 h beobachtet werden kann. In geringen Salzgehalten (5 ‰ S) tritt das erste Maximum verzögert ein (60 h) und liegt bei mittleren Werten von nur 175 g. Eidurchmesser und Eivolumen waren am größten in 5 ‰ S und lagen bei 20 und 23 ‰ S nahe den beobachteten Minima. Die Aushärtung der äußeren, klebrigen Eihülle war in geringen Salzgehalten verzögert. Auch die Inkubationszeit war durch den Salzgehalt beeinflußt. Sie war am längsten in 20 ‰ S (690–700 h) und am kürzesten in 35 ‰ S (545–570 h). Die Exposition der Heringseier gegenüber Cadmium veränderte alle beobachteten Parameter. In Abhängigkeit von der Cadmium-Konzentration verzögerte sich das Auftreten des ersten Druckfestigkeitsmaximums. Dieses war gleichzeitig erheblich kleiner: 200–350 g (1 ppm Cd) gegenüber 1300 g in den Kontrollen. Auch die Eivolumina nehmen mit steigender Cadmium-Konzentration ab.

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