

The effect of low pH on perch, *Perca fluviatilis* L. II. The effect of acid stress on different development stages of perch

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The pH sensitivity of perch eggs, fry, young of the year, and adult fishes was studied. The eggs were most sensitive soon after fertilization and fry after hatching while the adult perch were most tolerant. The variation in pH sensitivity of eggs between single females was wide. The pH sensitivity of eggs from two perch populations showed a difference at pH 4.0. The fry became acclimatised to the pH: the lower the pH at hatching, the better tolerance to low pH as fry. At a constant low pH there was a correlation between the conductivity of water and pH sensitivity of eggs.

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1. Introduction

The sensitivity of the earliest development stages of fish to low pH has been reported in many studies (Jensen & Snekvik 1972, Johansson et al. 1973, Almer et al. 1974, Beamish 1974, Harvey 1980), but only a few studies have tested the sensitivity of a fish species to low pH during its life span (Swarts et al. 1978, Edwards & Gjedrem 1979).

Differences in pH sensitivity between populations of a fish species have been reported by Gjedrem (1976), Robinson et al. (1976), Rahel & Magnuson (1980), Overrein et al. (1980). The role of synergistic factors, for example conductivity (Leivestad et al. 1976), aluminium (Muniz & Leivestad 1980) and other metals (Harvey et al. 1982), in the process of acidification is now an important object of research.

The present study concerns the differences in pH sensitivity of perch at different development stages, differences in acid tolerance of eggs from two populations of perch, and the effect of some properties of water on the mortality of perch eggs at low pH.

2. Material and methods

The field work was performed at Evo Inland Fisheries and Aquaculture Research Station, Lammi, southern Finland. Perch eggs used in the experiments were from three forest lakes, Lake Nimetön, Lake Valkea Mustajärvi, and Lake Horkkajärvi. They were fertilized in natural conditions and reared in one litre polyethylene jars. The water used was ground water from Evo Station with a natural pH of 6.4. The experimental pH values were adjusted with H₂SO₄ (analytical grade). Temperature was adjusted to fit the natural conditions with water from a near flowing river. It varied from 9.7°C at the beginning to 17.0°C at the end of the experiments.

2.1. The pH sensitivity of perch at different stages of development

Eggs from two females were used, one from Lake Nimetön and the other from Lake Valkea Mustajärvi. The eggs were kept in water from Evo Station and divided into clusters of 30-75 eggs. Four egg clusters from each female were transferred to pH 3.5, 4.0, 5.0, and 6.4 as follows: the first clusters ($n = 480$) were transferred immediately on arrival at the laboratory (stage I). They had developed for about 15 day degrees after spawning. The second clusters ($n = 330$) were transferred after 80 day degrees. These embryos had eyes, but no pigments, and showed some signs of movement (stage II). The third clusters ($n = 180$) were transferred after 120 day degrees. The embryos were well developed, pigmented and moved actively (stage III). The eggs of the fourth clusters hatched in control water after 150-180 day degrees and the pH sensitivity of the fry ($n = 160$) was tested at the same pH values (stage IV). The experiment was continued later in the summer when pH sensitivity of two month old young (stage V) and adult perch (stage VI) was tested. Values of pH and water used were the same as before. The fishes were kept in 150 l basins. All fish were fed with zooplankton, the adults also with benthic animals.

There were 25 young of the year per basin (mean length 31 mm, *SD* 2.1 mm) and 15 adults (mean length 108 mm, *SD* 17 mm) both caught from Lake Majajärvi, a small, very humic lake with water quality equal to that in Lake Nimetön. The length of the experiment was 3-16 days (50-250 day degrees). The developing eggs were reared to hatching: stage I at 240, II at 160 and III at 110 day degrees. The rearing time was 50 day degrees for fry and 250 for fingerlings and adults, corresponding to that of fertilized eggs at pH 4.0.

2.2. The pH sensitivity of perch eggs from different lakes

Roe of three females from Lake Valkea Mustajärvi and three from Lake Nimetön was used. The natural pH of the lakes was 6.4 and 5.8, respectively, in spring 1981. The roe was divided into clusters of 50-100 eggs and they were reared at pH 3.5, 4.0, 5.0 and 6.4 ($n = 150-280$ eggs at each pH from both lakes) so that eggs from 6 females were reared at every experimental pH. Dead eggs were counted daily, the eggs

remaining constantly under water, but were not removed in order to avoid damage to the live eggs. The difference in pH sensitivity of the eggs between the two populations was tested by binomial *t*-test for comparison of experimental percentage values (Mäkinen 1974). The same test was also used in the comparison of pH sensitivity of different stages of development of perch.

2.3. The effect of synergistic factors

In spring 1982 the effect of water quality on the pH sensitivity of perch eggs was tested by rearing eggs in different waters with constant pH. The eggs used here were from three females from Lake Horkkajärvi, a very humic lake of 1.1 ha and with water properties equal to those of Lake Nimetön. The rearing waters were tapwater from Lammi Biological Station and from Evo Station and lake water from Lake Karhujärvi and Lake Valkea Mustajärvi. The pH of each lot of water was adjusted to 4.0 with analytical grade H_2SO_4 . The electric conductivity and the concentrations of Na, K, Ca, Mg, Fe, and Al were compared with the mortality of the eggs by linear regression. From each female 50–70 eggs (170–200 eggs in each lot of water) were reared and the dead eggs counted daily. The control experiment was performed by rearing a corresponding number of eggs from the same females in the waters from all sources as such.

3. Results and discussion

3.1. The pH sensitivity of perch at different stages of development

The eggs of perch were most sensitive to low pH soon after fertilization (Fig. 1 I). The pH sensitivity decreased when the time between fertilization and transfer to low pH increased, so that development stage III was most tolerant. However, the fry were most sensitive immediately after hatching (Fig. 1 IV). They showed the highest mortality ($P < 0.001$) at pH 4.0 and 5.0. At pH 3.5 the mortality of fry was also highest ($P < 0.001$) after 10 and 50 day degrees, but later all eggs, stages I–III, died. At the control pH (6.4) no eggs or fry died. At pH 3.5 the stage I eggs showed higher mortality ($P < 0.001$) after 10 and 50 day degrees than those of stages II and III. The difference was not significant ($P > 0.1$) between stages II and III. At pH 4.0 the difference in the mortality of the eggs between stages I and II was significant after 10 day degrees ($P < 0.01$) and 50 day degrees ($P < 0.05$), but not at the end of the experiment ($P > 0.1$). The pH tolerance in stage III was higher than in I and II ($P < 0.01$). At pH 5.0 there were no significant differences in the pH sensitivity of the eggs from stages I–III.

The comparison of pH sensitivity between eggs, fry, young of the year and adult fishes showed that the pH tolerance of perch increases with the age of the fish: at pH 3.5 all the earliest development stages (I–IV) exhibited a 100 % mortality, but young of the year (stage V) exhibited 81 % mortality and the adults (stage VI) 47 %. It is important to note that this experiment was done

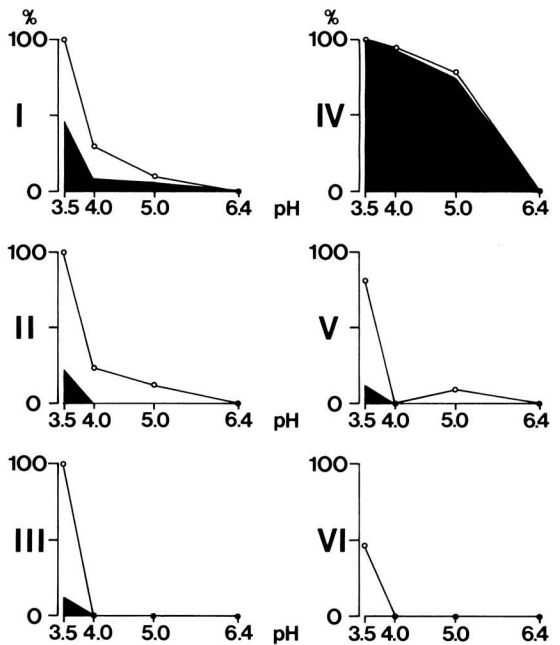


Fig. 1. The mortality (%) of perch eggs at different development stages (I–III), fry (IV), O+ young (V), and adult fishes (VI) at different pH after 10 day degrees (black) and at the end of the experiment (line).

without acclimatisation. Eggs, fry, fingerlings and adult perch were transferred to pH values different from that in the water of their own lake or from that in the control water (pH 6.4). In another experiment the fry showed acclimatisation: those hatched at pH 4.0 had a survival of 75 % at the same pH while those hatched at pH 6.4 had only 7 % survival at pH 4.0 after 50 day degrees (Table 1). The sharp changes in pH used in this work hardly occur under natural conditions. However, sudden decreases in pH of up to two pH units are recorded due to thaw waters in small lakes (Dickson 1980), which could affect the reproduction of perch.

The present results, showing that later transfer of the eggs to low pH yields a higher hatchig percentage agree with those of Edwards &

Table 1. The effect of low pH on perch fry hatched at different pH (survival % after 50 day degrees).

Eggs hatched at pH	Fry transferred to pH		
	3.5	4.0	5.0
4.0	0	75	100
5.0	0	25	50
6.4	0	7	22

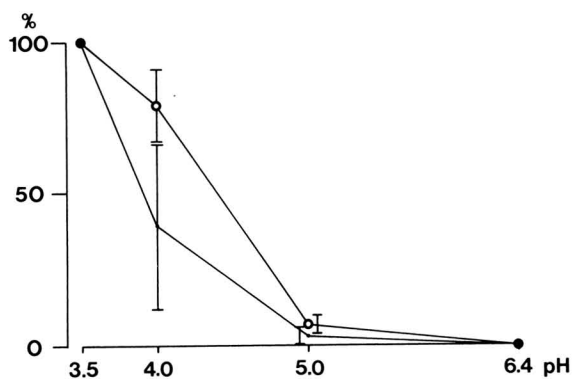


Fig. 2. The mortality (%) of perch eggs from Lake Valkea Mustajärvi (circles) and Lake Nimetön (dots). Vertical bars indicate S.E.

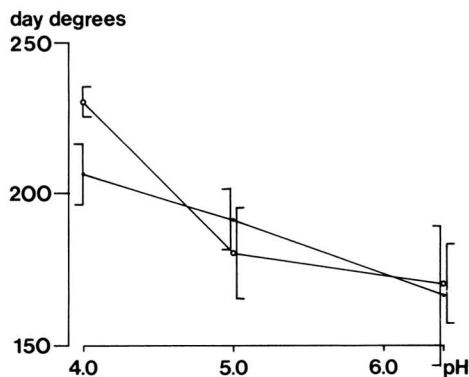


Fig. 3. The development time of perch eggs from Lake Valkea Mustajärvi (circles) and Lake Nimetön (dots) at different pH values. Vertical bars indicate S.E.

Gjedrem (1979) and Runn et al. (1977). In laboratory experiments it has been shown that fish eggs are most sensitive to low pH during their first hours after fertilization (Lee & Gerking 1980). The reason for this is the rapid hardening of the egg chorion after fertilization (Cykowska & Winnicki 1972). Thus, during the period of 15 hours between the fertilization of the eggs and the beginning of the experiment they may have already increased their tolerance to low pH. On the other hand, it has been suggested that the oogenesis of fishes is more sensitive to acidity than are eggs after spawning (Craig & Baksi 1977). The acidity has been reported to affect the reproductive physiology of maturing adults so that they do not spawn (Beamish et al. 1975). However, most observed cases of reproductive failures have been attributed to acid-induced mortality of eggs and fry (Schofield 1976, Fritz 1980), but with acute acid stress, adult fishes may die, too (Leivestad & Muniz 1976, Muniz et al. 1979).

3.2. The pH sensitivity of perch eggs from different lakes

The mortality of the eggs from Lake Valkea Mustajärvi and Lake Nimetön was equal at pH 3.5 (100%), 5.0 (7 and 3%), and 6.4 (0%) (Fig. 2). At pH 4.0 there was a difference ($P < 0.001$) in the egg mortality (79% in Lake Valkea Mustajärvi and 39% in Lake Nimetön). The variation in egg mortality at pH 4.0 between single females was wide. If this is due to differences in parent fishes, the reason might be genetic variations between the females or different general condition of the fishes during maturation of the eggs. The time between fertilization and the beginning of the experiment may also have varied by some hours,

which could have induced the different pH sensitivity. There was a difference in the increment of the development time of the eggs between the two populations at pH 4.0 (Fig. 3).

Genetic variation is one of the factors affecting the pH sensitivity of fishes (Gjedrem 1976, Grande et al. 1978). Crosses between brown trout strains in Norway have yielded higher survival rates than strain matings (Gjedrem 1980). Differences in the ability to resist acidic environments has also been demonstrated between strains of adult, juvenile, and embryonic brook trout (Swarts et al. 1978). Rahel & Magnuson (1980) showed a clear difference in pH sensitivity between two yellow perch populations. After an acclimatisation period of 21 days at pH 4.6 the pH tolerance of fishes from both populations increased, but the difference in tolerance between the populations remained.

3.3. The effect of synergistic factors

If perch eggs were reared in different waters (Table 2) at constant pH 4.0 the egg mortality and the conductivity of water showed a significant inverse correlation, $r^2 = 0.91$ (Fig. 4). The mortality of the eggs varied between 29 and 44% in the experiment and 3 and 11% in the control. The time course of the egg mortality (Fig. 5) showed that the difference in effect between the lake waters of low conductivity and the higher conductivity waters from Lammi and Evo Stations was greatest at the beginning of the experiment, but narrowed later. The cations Na, Ca and Mg showed a significant inverse correlation ($P < 0.05$) with the mortality of the eggs ($r^2 = 0.97, 0.92$ and 0.91 , respectively).

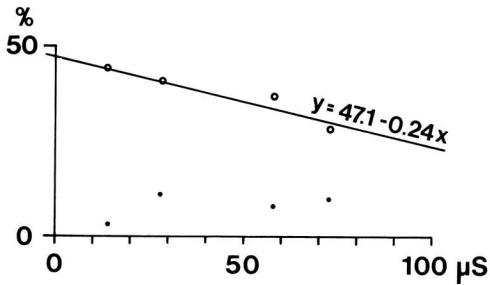


Fig. 4. The relation between egg mortality (%) and conductivity of water at pH 4.0 after 150 day degrees. Mortality at control also present (dots).

In southern Norway, for example, 70 % of recorded lakes of pH 4.9–5.1 and conductivity less than 10 μS have lost their fish populations, but less than 10 % of the lakes of the same pH and a conductivity of 30 μS or more (Muniz & Leivestad 1980). The disturbances in the salt balance of the fish are probably the reason for them dying at pH 4.0–5.0 (Fugelli & Vislie 1980). This is why increasing salt concentrations lower the lethal pH level (Leivestad et al. 1976). It may also explain why small fish are more sensitive to low pH than larger ones: they have greater body and gill surface areas per unit weight than larger fish, which allows more rapid ion fluxes because of the relatively greater exchange area in contact with the water (Robinson et al. 1976).

The aluminium concentration of the test waters showed a low correlation with the egg mortality.

Table 2. Properties of different rearing waters in testing the effect of synergistic factors (the values from the time of the experiment, except Na, K, Ca and Mg, which were analysed later).

	Tap water		Lake water	
	Lammi Station	Evo Station	Karhujärvi	Valkea Mustaj.
Natural pH	6.8	6.4	4.4	6.4
Conductivity ($\mu\text{S}/\text{cm}$ 20°C)	73	58	28	14
Na (mg/l)	4.1	2.0	1.1	0.9
K (mg/l)	1.5	1.7	0.9	0.7
Ca (mg/l)	13.2	8.3	2.7	2.6
Mg (mg/l)	4.1	1.1	0.8	0.5
Fe (mg/l)	<0.1	<0.1	0.2	0.2
Al ($\mu\text{g}/\text{l}$)	17	22	130	40

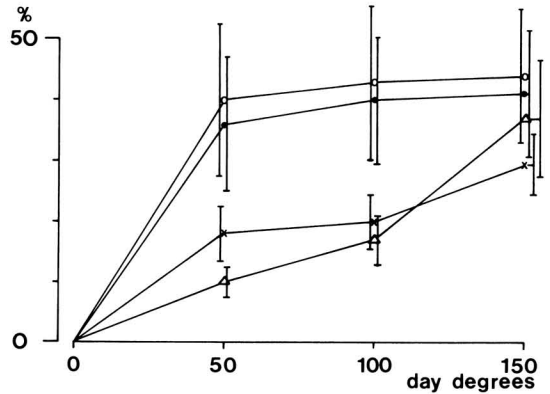


Fig. 5. The egg mortality (%) at pH 4.0 in water of Lammi Biological Station (crosses), Evo Station (triangles), Lake Karhujärvi (dots), and Lake Valkea Mustajärvi (circles). Vertical bars indicate S.E.

The reason may be the low aluminium concentrations of these waters, from 17 $\mu\text{g}/\text{l}$ to 130 $\mu\text{g}/\text{l}$, while concentrations of 200 $\mu\text{g}/\text{l}$ and more are reported to be toxic to fishes (Grahn 1980). The speciation of aluminium is shown to have a substantial effect on its toxicity to fish. Aluminium with no additional complexing agents is very toxic, but when complexed with organic compounds, its toxicity decreases (Baker & Schofield 1980). The test water with the highest aluminium concentration is extremely humic. This allows the possibility of humus-aluminium complexes, which may decrease the toxicity of aluminium.

There was no correlation between the low iron concentrations of the test waters (0.1–0.2 mg/l) and the egg mortality. Although there are reports of fish deaths in waters with high iron concentrations at different pH values (Vallin 1953), the significance of iron compounds as synergistic components with pH is not clear (EIFAC 1969). In addition iron compounds effectively precipitate out in natural waters due to humic substances (Ryhänen 1961).

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