NATIONAL SWEDISH BOARD OF FISHERIES

INSTITUTE OF FRESHWATER RESEARCH

DROTTNINGHOLM

Report No 61

LUND 1984
BLOMS BOKTRYCKERI AB

NATIONAL SWEDISH BOARD OF FISHERIES

INSTITUTE OF FRESHWATER RESEARCH

DROTTNINGHOLM

Report No 61

1984

Forword

The now virtually global aspects of the acidification problem were first realized in Norway. The impact of acid rain on the biological production of freshwater ecosystems has also been monitored for decades in Norway and Sweden. It is the intention of this volume of the Report series to focus attention on the multidisciplinary studies presently engaging researchers in the two countries, covering all aspects of freshwater ecosystems, from chemical and physical parameters to phytoplankton, zooplankton, benthic fauna and fish. The various effects of acidification are well-covered. The results of measures to counteract these effects, *viz.* liming, are also dealt with.

It is further hoped that the pooled knowledge displayed in this volume will serve as a platform for future environmental research relating to the acidification process, and also provide fresh arguments for administrators and politicians engaged in environmental issues.

Finally, on behalf of the Institute of Freshwater Research and all authors contributing to this volume, I would like to thank the National Environment Protection Board of Sweden for taking the initiative of funding the full production costs of this volume.

Lennart Nyman
Editor-in-Chief

ISSN 0082-0032

LUND 1984
BLOMS BOKTRYCKERI AB

Contents

Effects of Acidification on Age Class Composition in Arctic Char (*Salvelinus alpinus* (L.)) and Brown Trout (*Salmo trutta* L.) in a Coastal Area, SW Norway

RAGNVALD ANDERSEN,[1] IVAR P. MUNIZ,[2] and JOSTEIN SKURDAL [1]

[1] University of Oslo, Department of Biology, Division of Zoology, P.O. Box 1050, Blindern, N-0316 Oslo 3, Norway.

[2] University of Oslo, Department of Biology, Division of General Physiology, P.O. Box 1051, Blindern, N-0316 Oslo 3, Norway.

ABSTRACT

We have studied the Arctic char (*Salvelinus alpinus*) and the brown trout (*Salmo trutta*) in two small coastal watersheds in SW Norway during the years 1976—83. All the lakes and streams are markedly influenced by marine salts, and are acidified by approximately 0.5—1.5 pH units. Historically, the lakes in this area were often densely populated with small-sized Arctic char and brown trout. By 1983, the acidified headwater lakes had lost their fish stocks, while the remaining fish stocks were in various stages of local extinction. These stocks were either characterized by a dominance of old (ageing) or young fish (juvenilization). Both growth and quality are now good. Arctic char are more affected than brown trout and this is probably due to differences in tolerance between species.

The observed population responses are probably reflecting differences in habitat utilization between species and populations, particularly with respect to their spawning and nursery habitat. The Arctic char and one brown trout stock showed ageing, characterized by total or partial recruitment failure but with low mortality on postspawners. The brown trout also showed juvenilization with high postspawning mortality. We suggest that juvenilization is due to poor water quality during the spawning season.

I. INTRODUCTION

Deposition of airborne pollutants and acidification of freshwater has inflicted a major deterioration on Norwegian freshwater fish resources (JENSEN and SNEKVIK 1972, LEIVESTAD *et al.* 1976). Loss of fish is documented in a 33,000 km^2 area of southern Norway (SEVALDRUD and MUNIZ 1980). The decline started at the turn of the century, but was first noted as a major problem in the 1950's and 60's (JENSEN and SNEKVIK 1972).

The most severely affected region is our four southernmost counties (SEVALDRUD and MUNIZ 1980). The frequency of barren lakes increase with increasing acidity. Fish are lost at higher pH's in low-conductivity lakes. Fish loss started in headwater lakes and has over the years gradually spread downstream. Today, fish are mainly confined to larger lowland lakes with higher ion content (SEVALDRUD and MUNIZ 1980). Stocks in acid-stressed lakes are often diminished and in various stages of local extinction (HARVEY 1982). They are either characterized by a dominance of old (ageing) or young fish (juvenilization) (HARVEY 1980, ROSSELAND *et al.* 1980).

We have studied Arctic char (*Salvelinus alpinus*) and brown trout (*Salmo trutta*) in two small coastal watersheds in southwestern Norway during 1976—83. Based on present fish status and stock characteristics, we have compared these species with regard to their sensitivity to acidification and different modes of extinction, *i.e.* ageing or juvenilization. We have also compared the response of ecologically differentiated creek and lake spawning populations of brown trout.

II. STUDY AREA

Selura and Djupvikvatn watersheds are situated near Flekkefjord in the county of Vest-Agder, SW Norway (Fig. 1). The climate at Flekkefjord is oceanic, and the annual rainfall is high, both compared to the coastal metereological station Lista (25 km further south), and to the inland station Skreådalen (60 km further north). The

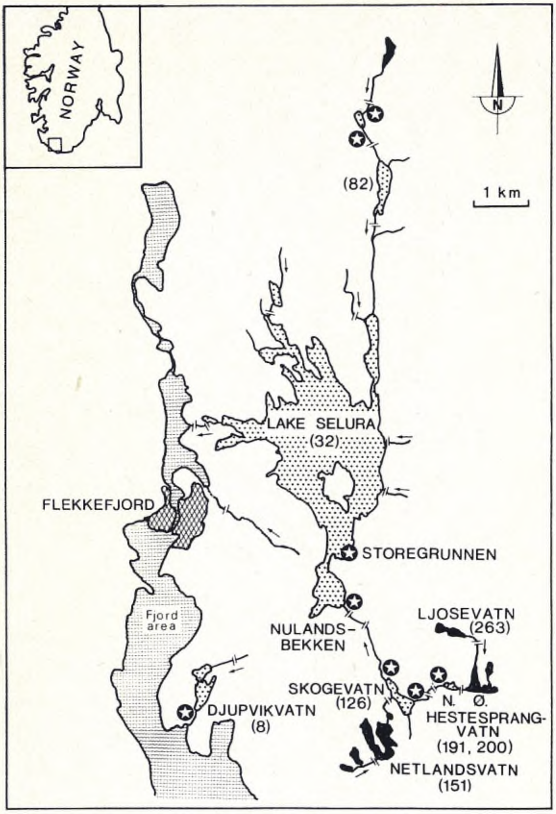


Fig. 1. The Selura and Djupvikvatn watersheds, SW Norway. Barren lakes are indicated in black and sites with current brown trout reproduction with stars. Altitudes above sea level (m) are in parentheses.

heaviest rainfall and subsequent deposition in this region occur during late autumn (Fig. 2). The mean annual wet deposition of excess sulphate for the years 1972—82 was 1.2 (0.9—1.5) g S/m² at both Lista and Skreådalen. At Lista and Flekkefjord snow accumulation was small due to mild winters with alternating periods of melting and deposition (Fig. 2). At Skreådalen a substantial snow accumulation occurred throughout winter with a maximum snow depth of about 80 cm and snow melt in late April and early May.

The geology of the area is predominately banded and granitic gneisses (FALKUM 1972) with small glacifluvial deposits (ANDERSEN 1960).

The vegetation is typical for these coastal areas, *i.e.* a mixture of deciduous and coniferous forest in lower parts, with moors and bare rocks in the upper parts of the watersheds.

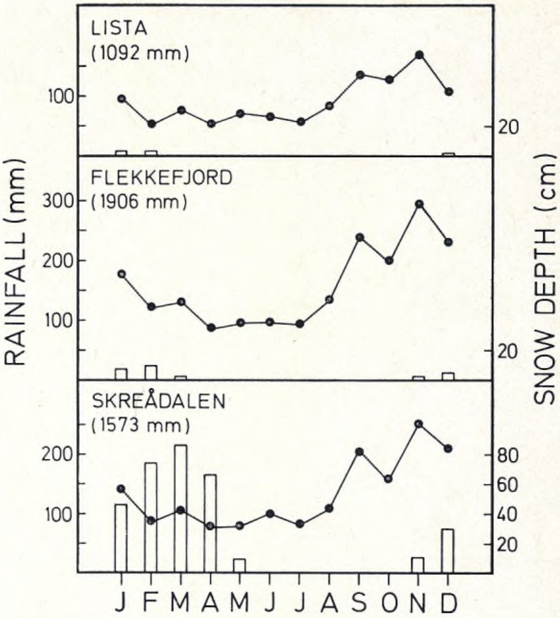


Fig. 2. Mean monthly precipitation (1972—82) and snow depth (columns) at the metereological stations Lista, Flekkefjord and Skreådalen, SW Norway. Mean yearly precipitation amounts in parentheses.

Both watersheds are sparsely populated and the 3 % of arable land area is mainly concentrated around Lake Selura.

The Selura watershed is 45 km², and includes 15 lakes and tarns. Lake Selura (510 hectares) which is the largest of the lakes has a maximum depth of 65 m. Netlandsvatn* (25 hectares) and Skogevatn (14 hectares) are more shallow lakes (maximum depths 25—20 m) and drains into the southern bight of Lake Selura (Fig. 1).

The other watershed, Djupvikvatn is 2.4 km² and has only one small lake, Djupvikvatn (18 hectares) with a maximum depth of 30 m, and drains directly into the sea.

III. MATERIAL AND METHODS

We have studied the fish communities of Lake Selura, Skogevatn, and Djupvikvatn. The lakes were fished in the autumn and representative samples were obtained by using chains or single bottom nets (1.5 m×25 m, mesh sizes: 10—

---

* The suffix "vatn" means lake.

45 mm), a beach seine (mesh size: 9 mm), hoop nets (mesh size: 13 mm) and by electrofishing the tributaries. Catch per unit effort (CPUE) is the number of fish caught during a 12 hr period in nets with mesh sizes; 19.5, 22.5, 26, 29, 35, 39 and 45 mm. These mesh sizes catch brown trout from 18 to 40 cm with approximately equal efficiency (JENSEN 1977).

Our fish material from the southern bight of Lake Selura consists of 187 Arctic char and 125 brown trout. An additional 636 brown trout were caught at their spawning area in the lake (Storegrunnen) and 1076 brown trout during their spawning run in the creek (Nulandsbekken) (Fig. 1). The material from Djupvikvatn and Skogevatn consists of 2 Arctic char, 121 brown trout and 110 Arctic char, 150 brown trout, respectively. 36 of the Arctic char and 51 of the brown trout from Skogevatn caught in the summer of 1982 were not used for calculating CPUE.

In the autumn of 1982 and 1983 we surveyed all the major brooks by electroshocking and captured 83 fry and parr and 19 mature brown trout.

Natural tip length (RICKER 1979) was recorded to the nearest millimetre and weight to the nearest gram. Sex and stage of sexual maturity were determined according to DAHL (1917) and the fish were aged from otoliths (NORDENG 1961, JONSSON 1976). The Arctic char otoliths were burnt and sectioned before reading (CHRISTENSEN 1964, BLACKER 1974). Fish lacking annuli in their otoliths were assigned to age-group 0, those with one annulus to age-group 1, etc. .

The rate of survival (S) was estimated from age-allocated CPUE data in successive years (JACKSON 1939, RICKER 1975).

Growth curves of the von Bertalanffy type were fitted to the data using ALLEN's (1966) method for obtaining the best least-squares estimates of the parameters L_∞, K and T_0 in the equation $L = L_\infty\ (1\text{-}e^{-k(T-T_0)})$. Differences between the various parameters were tested by Student's t-test. The length-weight relationship was calculated from: $\text{Log}\ W = \log a + b \log L$, and the condition factor from: $K = 10^5 \cdot a \cdot L^{b-3}$, where W is the weight in grams, L the length in millimetres, and a and b are constants.

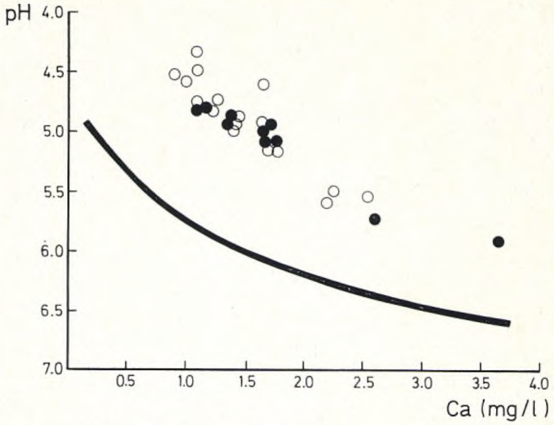


Fig. 3. The relationship between pH and calcium concentration for lakes (●) and streams (○) in the study area, with HENRIKSEN's (1979) "acidification indicator" line. The lake samples were taken at surface (1 m) and 1 m above the bottom.

Water samples were taken from major lakes and tributaries in the watersheds in late October 1983. pH and conductivity were measured in the field and aliquots were prepared to determine acid reactive Al and non-labile monomeric Al, using DRISCOLL's fractionation procedure (DRISCOLL 1980). The aliquots were frozen and later analyzed by the catechol-violet method (WRIGHT and SKOGHEIM 1983). The other chemical constituents were measured in the laboratory using standard methods.

IV. RESULTS

Water quality

Lakes and brooks in the area are oligotrophic, low in colour (< 10 mg Pt/l) and have practically no alkalinity. The water is markedly influenced by marine salts (Na: 6.7—9.7 mg/l; Cl: 9.0—16.4 mg/l) and the conductivity is relatively high (20: 37—67 µs/cm). If we plot pH against Ca (Fig. 3) and use HENRIKSEN's (1979) acidification concept and his nomogram, all our data points lie above the line of full alkalinity. This indicates that all lakes and streams at present are acidified by approximately 0.5—1.5 pH-units. The concentration of toxic labile monomeric Al increases

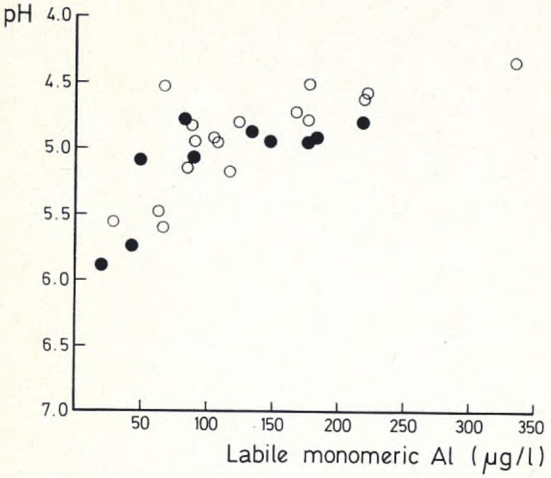


Fig. 4. The relationship between pH and concentration of labile monomeric aluminium in lakes (●) and streams (o) in the study area, for the same samples as in Fig. 3.

with increasing acidity (Fig. 4). This is a common property of acidfied water (DICKSON 1980, DRISCOLL 1980).

Fish status

The lakes of the study area had previously good, often dense, selfsustaining stocks of Arctic char and brown trout. Arctic char were found in Netlandsvatn, Skogevatn, Lake Selura and Djupvikvatn, while all lakes harboured brown trout. Arctic char were typically stunted, whereas brown trout were more variable.

In the late 1950's landowners registered a drop in their catches in the headwater lakes, Ljosevatn and Ø. Hestesprangvatn, and several of the headwater lakes draining into Nulandsbekken are now devoid of fish. Naturally reproducing brown trout is at present found in N. Hestesprangvatn, Skogevatn, Lake Selura and Djupvikvatn (Fig. 1). All streams with a labile monomeric Al content ≥ 125 µg/l in October 1983 were found to be barren in both 1982 and 1983 surveys. The density of Arctic char and brown trout is highest in Lake Selura (Table 1). Arctic char and brown trout undergo habitat segregation during the summer period of thermal stratification and CPUE for Arctic char therefore refers to fish captured in chains of nets extending from the sublittoral into the profundal zone. CPUE for brown trout refers to fish from single nets in shallow water. The catch of brown trout was about equal in Skogevatn and Djupvikvatn, whereas Arctic char were more abundant in Skogevatn.

Table 1. *CPUE for Arctic char and brown trout.*

Locality	Year	Effort	Arctic char	Brown trout
Skogevatn	1982—83	4	15.8	19.0
L. Selura	1982	4	30.3	—
	1983	2	—	61.5
Djupvikvatn	1982—83	7	0.3	12.6

Age-class structure and length distribution

In 1976, the Lake Selura Arctic char were stunted with a maximum length of 21 cm (Fig. 5). In 1982, the maximum length had increased to 31 cm with 57 % of the Arctic char exceeding 21 cm. Changes in age composition during the years 1976—82 shows that the stock had become more aged. Only 4 % of the Arctic char were older than 10 years in 1976 compared to 42 % in 1982, and the maximum age increased from 12 to 16 years (Fig. 5).

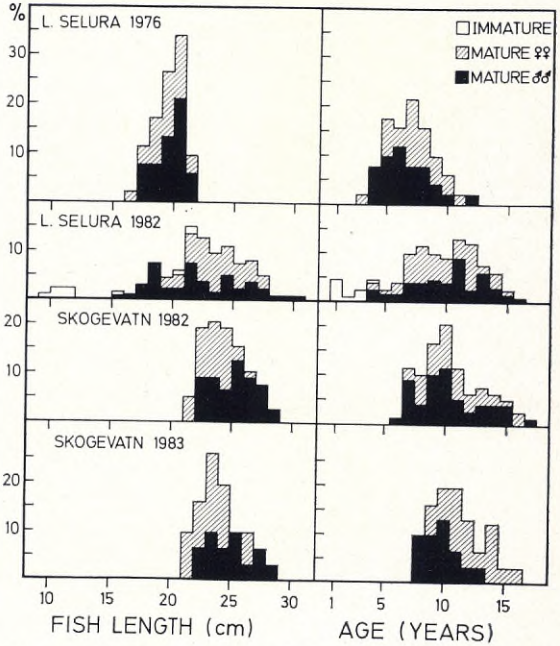


Fig. 5. Length and age distribution (in %) of Arctic char sampled from Lake Selura and Skogevatn.

The length distribution of the Skogevatn Arctic char in 1982 and 1983 was unimodal covering the length interval 21—29 cm with no apparent changes (Fig. 5). The smallest individuals of the stock were thus equal to the largest individuals in Lake Selura in 1976. The stock was dominated by older fish and suffered from total recruitment failure. Younger year-classes were missing, 1—5 in 1982 and 1—7 in 1983. Survival rate (S) from 1982 to 1983 was estimated to 0.79.

The Djupvikvatn Arctic char was nearly extinct and the two specimens caught in 1982 were both old (9 and 14 years).

In 1977—78, there was a significantly higher fraction of older fish among the lake spawning compared to the creek spawning brown trout in Lake Selura ($\chi^2 = 337.3$, $P < 0.001$). The oldest creek spawner was 6 years, while some lake spawners even exceeded 10 years (Fig. 6). In both populations first time spawners were 1^+ among males and 2^+ among females.

In the spawning season of 1978, both populations were heavily fished with gill and hoop nets and by electroshocking in the creek Nulandsbekken. By using mark-recapture techniques it was estimated that the number of mature fish in both populations was reduced by 80 %. The two populations responded quite differently to this manipulation. In 1982, the creek spawners showed a weak 1979 year class (spawned in 1978) but had otherwise recovered with a similar age-class composition as in 1977—78 (Fig. 6). The lake spawners, however, showed no distinct year-class fluctuations, but in 1982 the population consisted mainly of 1—4 year-old fish (*i.e.* year classes 1978—81). The small fraction of older fish (*i.e.* year classes 1977 and below) most probably was a direct effect of the large population reduction in 1978.

In 1977—78, 19 % of the lake spawners were larger than 25.0 cm and the largest fish measured 37 cm, whereas the corresponding figures for 1982 were 10 % and 39 cm. Among the creek spawners we found that only 3 % exeeded 25.0 cm with a maximum length of 34 cm in 1977—78, while the largest fish in 1982 measured 35 cm. In that year, however, we found that 12 % of the creek spawners were larger than 25.0 cm which can be attributed to increased growth.

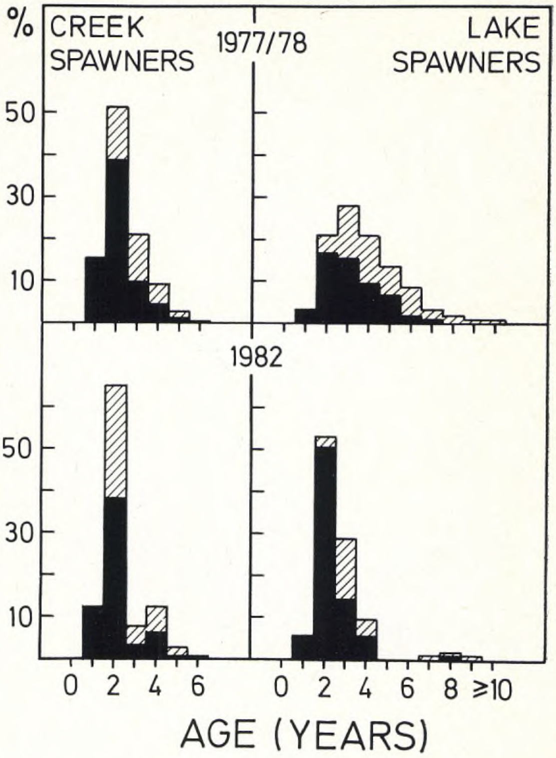


Fig. 6. Age distribution (in %) of sexually mature lake and creek spawning brown trout captured in Lake Selura. (Same legends as in Fig. 5.)

Brown trout in Skogevatn were in 1982 dominated by young fish, age classes 1—3, the brown trout in Djupvikvatn by age classes 3—6 (Fig. 7). In 1983, several of the oldest age groups in Skogevatn were much reduced or missing. This may have been caused by heavy mortality on mature as well as immature fish (cf. Fig. 7). The brown trout in Djupvikvatn seems to move in the opposite direction. Age classes 4—7 were the most frequent classes of older fish in 1983 and there were also considerable year-class fluctuations. That year, age class 2 accounted for more than 35 % of the sample. Survival rates of fish older than 2 years were 0.38 for the Skogevatn brown trout and 0.81 for the Djupvikvatn brown trout.

The brown trout males in both these lakes reached sexual maturity in their second year (age-group 1) while females in Skogevatn matured at age group 2, and at age group 3 in Djupvikvatn (Fig. 7).

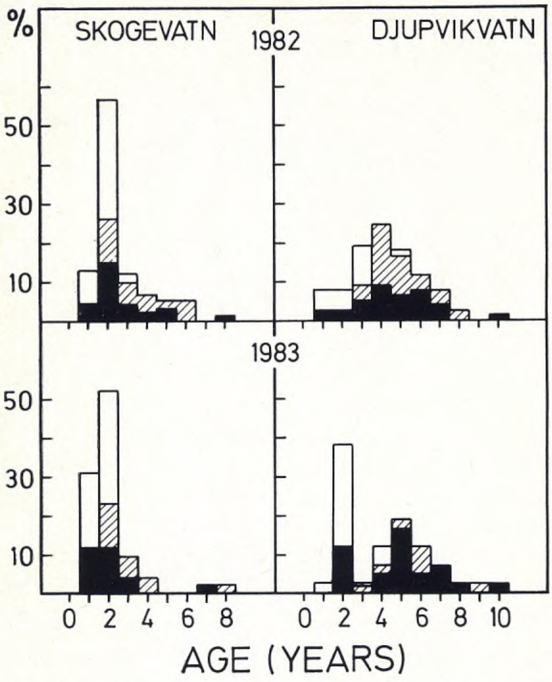


Fig. 7. Age distribution (in %) of brown trout sampled from Skogevatn and Djupvikvatn. (Same legends as in Fig. 5.)

The brown trout in Djupvikvatn attained a relatively large size. In 1982, 66 % were larger than 25.0 cm while 45 % exeeded this length in 1983. The brown trout in Skogevatn were smaller in size and in 1982, only 17 %, and in 1983, 10 % of the brown trout were larger than 25 cm.

Growth

A comparison of the growth of Arctic char in Lake Selura revealed that L_∞ was significantly higher ($P < 0.001$) and K lower ($P < 0.05$) in 1982 compared to 1976 (Table 2. In 1982, L_∞ was 242 mm for both the Arctic char from Lake Selura and Skogevatn.

There were no significant differences in growth between the creek and lake spawning brown trout in Lake Selura in 1978. The creek spawners had similar growth curves in 1978 and 1983, but the lake spawners showed significant difference in L_∞ ($P < 0.05$) and K ($P < 0.01$) between the years 1978 and 1982. The growth curves for the brown trout in Skogevatn and Djupvikvatn did not change significantly between the years 1982 and 1983.

The length-weight relationship

There were no significant differences in the length-weight relationship between immature and mature males and females of both species and therefore we have pooled such data. All regressions were highly significant and the coefficients of determination were 0.80—0.90 for the Arctic char and 0.98—0.99 for the brown trout (Table 3). The b-value for the Arctic char in Lake Selura increased significantly from 2.026 in 1976 to 3.202 in 1982 ($t=4.62$, $P < 0.001$), but the b-value for 1982 was not significantly different from 3.0 which corresponds to isometric growth. Thus, while the condition factors decreased markedly

Table 2. *Growth curves of the von Bertalanffy type for Arctic char and brown trout (L_∞= the mathematical asymptote of the curve (mm), K=a measure of the rate at which the growth curve approaches the asymptote, T_0=hypothetical starting time. Standard errors are in parentheses).*

Locality	Year	N	L_∞	K	T_0
Arctic char					
L. Selura	1976	51	196 (2)	1.606 (0.588)	1.89 (0.48)
	1982	103	242 (3)	0.611 (0.109)	0.09 (0.21)
Skogevatn	1983	31	242 (6)	1.041 (0.246)	6.88 (0.30)
Brown trout					
L. Selura, lake spawners	1978	275	343 (13)	0.246 (0.031)	−1.14 (0.25)
	1982	126	575 (138)	0.098 (0.042)	−2.15 (0.62)
L. Selura, creek spawners	1978	546	358 (25)	0.229 (0.037)	−1.20 (0.19)
	1983	91	464 (142)	0.162 (0.088)	−1.15 (0.45)
Skogevatn	1983	52	275 (13)	0.526 (0.122)	−0.89 (0.35)
Djupvikvatn	1983	42	288 (11)	0.771 (0.297)	0.21 (0.55)

Table 3. *Length — weight relationship in Arctic char and brown trout, with 95 % confidence intervals, and calculated condition factor (K̂, at fish length 200, 250 and 300 mm).*

Locality	Year	Number of fish	b	Log a	Coefficient of determination	F-value of variance of regression	K̂ 200	250	300
Arctic char									
L. Selura	1976	21	2.026 (1.554—2.498)	−2.831 (−2.890—−2.772)	0.80	79.4***	0.85	0.68	
	1982	95	3.204 (2.987—3.442)	−5.535 (−5.623—−5.477)	0.90	867.4***	0.86	0.90	
Skogevatn	1983	31	3.607 (2.967—4.245)	−6.519 (−6.630—−6.408)	0.82	132.9***	0.75	0.86	
Brown trout									
L. Selura,	1978	275	2.992 (2.948—3.038)	−4.985 (−5.052—−4.918)	0.98	1665.0***	0.99		0.99
lake spawners	1982	57	3.044 (2.946—3.142)	−5.100 (−5.154—−5.046)	0.98	384.8***	1.00		1.02
L. Selura,	1978	546	2.887 (2.856—2.918)	−4.754 (−4.814—−4.694)	0.98	3342.0***	0.97		0.92
creek spawners	1983	87	2.962 (2.896—3.028)	−4.869 (−4.933—−4.805)	0.99	807.4***	1.11		1.09
Skogevatn	1983	52	3.209 (3.072—3.346)	−5.465 (−5.529—−5.401)	0.98	220.3***	1.04		1.13
Djupvikvatn	1983	42	3.034 (2.944—3.124)	−5.086 (−5.139—−5.033)	0.99	459.8***	0.98		1.00

Significance; ***, $P \leq 0.001$

with length in 1976 the variation with length was small in 1982.

The b-value of the Arctic char in Skogevatn was about 3.6 both in 1982 and 1983, but our testing revealed no significant difference from isometric growth. The calculated condition factor of Arctic char in our study lakes was 0.68—0.84 in 1976 and 0.75—0.90 in 1982—83.

In the creek-spawning brown trout in Lake Selura, b increased from 2.887 in 1978 to 2.962 in 1982 ($t=2.17$, $P < 0.05$). In the lake spawners, b was about 3.0 in both years. There were significant differences in the b-value between the populations in 1978 ($t=3.77$, $P < 0.001$) but not in 1982. The b-value of the brown trout in Skogevatn in 1982 and Djupvikvatn in both 1982 and 1983, was about 3.0. In 1983, the b-value for Skogevatn was significantly higher than 3.0. The calculated condition factor of the brown trout in these lakes was 0.92—0.99 in 1978 and 0.98—1.13 in 1982/83.

V. DISCUSSION

In the early 1970's, the inland lakes in southernmost Norway had experienced considerable losses of fish (Jensen and Snekvik 1972). At that time, fish were still present in all our study lakes (Snekvik 1974, Sevaldrud and Muniz 1980). This timelag is probably related to higher content of dissolved salts in the water of coastal lakes (Leivestad *et al.* 1976), and also to differences in snow accumulation between inland and coastal localities. At inland localities snow accumulates throughout late autumn and winter (cf. Fig. 2). During spring thaw those pollutants which accumulate in the snow pack leach out in high concentrations, and such melting episodes are critical for fish reproduction and survival (Henriksen and Wright 1977, Dickson 1980). At coastal sites with mild winters, only a small fraction of the deposition accumulates in the snow pack and fish will as a rule therefore not experience major spring thaw episodes.

At present (1983), loss of fishes within the Lake Selura watershed is confined to small headwater lakes situated at altitudes above 150 m. Lake N. Hestesprangvatn (195 m) still harbours a sparse stock of brown trout. This is probably associated with a temporal improvement in water quality due to a recent road construction along the southern shoreline. The road construction also may have had a positive influence on the Skogevatn brown trout, whose spawning now is restricted to the inlet brook from N. Hestesprangvatn and to the lake outlet. The density of Arctic char and brown trout in Skogevatn is low. Both Arctic char and brown trout are more abundant in the larger Lake Selura, further downstream. In the neighbouring Djupvikvatn both Arctic char and brown trout are strongly affected. The stock of Arctic char is close to extinction with only a few old individuals left.

Our study area is relatively small, and therefore probably experiences only small differences in precipitation amounts and acid loading. In spite of the coastal proximity of Djupvikvatn there are only minor differences in marine influence. The observed responses in the fish stocks in our area therefore are reflecting differences in reactivity between the various watersheds. Vegetation and soil are poorly developed around the headwater lakes, while the areas below have deeper soils and more lush vegetation. The observed patterns of local extinction in the study area are in agreement with the general observation that loss of fish stocks begins in small headwater lakes with a gradual spread downstream (SEVALDRUD and MUNIZ 1980).

In 1976, the Arctic char in Lake Selura were stunted and none exceeded 21 cm. The quality was poor and the condition factor decreased markedly with length. During subsequent years there has been a significant increase in both condition factor and growth. At present more than 50 % of the Arctic char exceed 21 cm. The improvement in growth and quality in acidifying lakes is probably due to changes in inter- and intraspecific competition (JENSEN and SNEKVIK 1972, ROSSELAND *et al.* 1980, RYAN and HARVEY 1980).

We used a beach seine in 1976, but because immature Arctic char often occupy deep benthic and pelagic habitats (HINDAR and JONSSON 1982) they are largely lacking in our 1976 material. The data from 1976 and 1982 are therefore not strictly comparable with respect to parr and immature fish. In Lake Selura, the fraction of Arctic char older than 10 years increased from 4 % to 40 % in the years 1976 to 1982. In 1982, age-group 4—6 became less abundant which may indicate partial recruitment failure.

The Arctic char in Skogevatn suffer from permanent recruitment failure. The fraction of old individuals is high and the estimated rate of survival is 0.79. Thus, in spite of recruitment failure the survival rates of mature char in both lakes are high.

There have been only minor changes in the brown trout populations in Lake Selura. The changes in growth and length-weight relationship in the lake spawning brown trout are probably caused by the 80 % population-reduction in 1978, and the fraction of old fish is low. The creek spawners had almost the same age-class composition in 1982 as in 1977—78. The 80 % population reduction resulted in a weak 1979 year-class of creek spawners, but had little influence on year-class strength among the lake spawners. This may indicate that egg and fry mortality is higher among the creek spawners.

There were considerable year-class fluctuations in the brown trout from Djupvikvatn with a strong age group 2 in 1983, but the mature fish seem to have become more aged. This stock response is probably a result of partial recruitment failure with intermittent reproduction. Survival rate in mature brown trout was 0.81, which is high and at the same level as for Arctic char.

Based on tagging-recapture (1977—78) experiments, the instantaneous rate of natural mortality (M) has been estimated to be 0.77 for lake and 0.96 for creek spawners in Lake Selura (ANDERSEN unpubl.). This is an exceptionally high post-spawning mortality, particularly for the creek spawners, and generally much higher than compared to data from unaffected localities in Norway (JENSEN 1972 and 1977). This may be an effect of critical water quality during the heavy autumn rains, in the spawning season, when the mature brown trout migrate into streams. Data on age-class composition from the creek spawners in Lake Selura also shows that they were more affected than the lake spawners (Fig. 6).

Our data on CPUE, age structure and recruitment as well as growth and condition factors indicate that Arctic char are more affected than brown trout. The two species utilize different habitats during their lifetime. Arctic char is generally a lake spawner and utilizes the lake as nursery and feeding areas. During the period of summer stagnation, Arctic char are often restricted to the hypolimnion (ANDERSEN and NILSSEN 1984). Brown trout spawn in running waters, though lake spawning may occur, and their nursery and feeding areas are generally in the brooks and the littoral zone. Therefore the Arctic char is better protected against rapid fluctuations in water quality. The observed differences in stock structure probably reflects species-specific differences in tolerance found in field tests (BUA and SNEKVIK 1972). ALMER *et al.* (1974) compared lake pH with test-fishing data and information on historical changes, and concluded that Arctic char was the more sensitive species. Our data indicate that egg and fry are the more sensitive stages in Arctic char because survival in mature stages is high in the acid-stressed lakes.

Our data demonstrate two different types of population responses, namely ageing and juvenilization. Ageing, as shown by the Arctic char in our lakes and for the brown trout in Djupvikvatn, is characterized by: (1) missing year classes, (2) dominance of repeat spawners, and (3) old and large fish. This is due to partial or total recruitment failure caused by increased mortality on eggs and fry, and increased survival of mature fish. Ageing is observed for lake spawning fish such as Arctic char, roach (*Rutilus rutilus*) and perch (*Perca fluviatilis*) (ALMER 1972, Almer *et al.* 1974, ROSSELAND *et al.* 1980, RYAN and HARVEY 1980), but was observed also in the outlet-spawning brown trout stock in Djupvikvatn. Fluctuations in recruitment are typical in such situations and may reflect year to year variation in water quality (HULTBERG 1976). The snow melt periods in localities with cold winter affect recruitment strongly and thus population ageing is often seen in such localities.

Juvenilization as shown by the brown trout from Lake Selura (creek spawners) and Skogevatn is characterized by: (1) lack of repeat spawners, (2) low age at sexual maturity, and (3) young and smaller fish. A combined effect of increased mortality on post-spawners and on eggs and fry, probably leads to juvenilization. Depending on the spawning and nursing habitat egg and fry mortality may have different effects on recruitment. If the spawning area is limited, any increase in egg mortality will be accompanied by a reduction in recruitment. If, on the other hand, the nursery habitat is limited, then recruitment can still be maintained at the same level as long as the number of fry produced is larger than the carrying capacity of the nursery area (cf. MORTENSEN 1977). Juvenilization was observed previously among creek spawners (ALMER 1972, BEAMISH *et al.* 1975, ROSSELAND *et al.* 1980). In lowland lakes autumn rain influences water quality directly (HARVEY 1980), and this may coincide with the spawning season of brown trout. The rainfall in Flekkefjord in the months October—December in 1976 and 1977 was 590 and 915 mm, respectively. The instantaneous natural mortality for the creek spawners in Lake Selura was 0.59 and 0.96, respectively in these years (ANDERSEN unpubl.). This shows an extremely high mortality in spawners in years with heavy rainfall during the spawning season, and we therefore suggest that juvenilization is an effect of poor water quality during the spawning season.

VI. ACKNOWLEDGMENTS

We are grateful to MIKAEL THUNESTVEDT and TOR NULAND for valuable help during the field work, and to HANS NORDENG for stimulating discussion. Water samples were analysed by ODD SKOGHEIM at The Directorate for Wildlife and Freshwater Fish, Norway. The Norwegian Meteorological Institute provided data on rain fall and snow depth and Norwegian Institute for Air Research (NILU) data on sulphur deposition. We are grateful to H. H. HARVEY and P. NYBERG for valuable comments. LISS FUSDAHL and RAGNHILD FRILSETH typed the manuscript. We are indebted to them all. Financial support for this research has been provided from Zoological Institute of the University of Oslo, Directorate for Wildlife and Freshwater Fish, Norway, Professor Robert Collett's bequest and Norwegian Council for Scientific and Industrial Research (NTNF).

VII. REFERENCES

Allen, K. R. 1966. A method of fitting growth curves of the von Bertalanffy type to observed data. J. Fish. Res. Board Can. 23: 163—179.

Almer, B. 1972. Effects of acidification of fish populations in lakes on the Swedish west coast. Inform. Inst. Freshw. Res., Drottningholm (12). 47 p. (In Swedish with English summary.)

— W. Dickson, C. Ekström, E. Hörnström and W. Miller. 1974. Effects of acidification on Swedish lakes. Ambio 3: 30—36.

Andersen, B. G. 1960. "Sørlandet" in late and postglacial time. Norges Geol. Undersøk. NGU 210: 1—142.

Andersen, R. and J. P. Nilssen. 1984. Mechanisms of co-existence of *Heterocope saliens* (Crustacea, Copepoda) and planktivorous fish. Fauna norv. Ser. A. (In the press.)

Beamish, R. J., W. L. Lockhart, J. C. Van Loon and H. H. Harvey. 1975. Long term acidification of a lake and resulting effects of fishes. Ambio 4: 98—102.

Blacker, R. W. 1974. Recent advances in otolith studies. p. 67—90. *In* Sea Fisheries research. Ed.: F. R. Harden Jones. Elek Science, London.

Bua, B. and E. Snekvik. 1972. Hatching experiments with eggs from salmonid fishes 1966—1971. Effects of acidity and dissolved salt in the water. Vann 7: 86—93 (In Norwegian.)

Christensen, J. M. 1964. Burning of otoliths, a technique for age determinations of soles and other fish. J. Cons. int. Explor. Mer. 29: 73—81.

Dahl, K. 1917. Studies and experiments on brown trout and brown trout lakes. Centraltrykkeriet, Kristiania. 107 p. (In Norwegian.)

Dickson, W. T. 1980. Properties of acidified waters p. 75—83. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

Driscoll, C. T. 1980. Chemical characterization of some dilute acidified lakes and streams in the Adirondack Region of New York State. Ph. D. Thesis. Cornell Univ., Ithaca, N. Y. 308 p.

Falkum, T. 1972. Geological map of the Flekkefjord area, Vest-Agder, South Norway. M: 100 000. NGU, Geofysisk avd.

Harvey, H. H. 1980. Widespread and diverse changes in the biota of North American lakes and rivers coincident with acidification. p. 93—98. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway. 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

— 1982. Population responses of fishes in acidified waters. p. 227—241. *In* "Acid rain/Fisheries". Proc. Int. Symp. Acidic Precip. Fish. Impacts Northeastern North America. Ed.: R. E. Johnson. American Fisheries Society.

Henriksen, A. 1979. A simple approach for identifying and measuring acidification in freshwater. Nature, Lond. 278: 542—544.

— and R. F. Wright. 1977. Effects of acid precipitation on a small acid lake in Southern Norway. Nord. Hydrol. 8: 1—10.

Hindar, K. and B. Jonsson. 1982. Habitat and food segregation of dwarf and normal Arctic char (*Salvelinus alpinus*) from Vangsvatnet Lake, Western Norway. Can. J. Fish. Aquat. Sci. 39: 1030—1045.

Hultberg, H. 1976. Thermally stratified acid water in late winter — a key factor inducing self-accelerating processes which increase acidification. Wat., Air, Soil Poll. 7: 279—294.

Jackson, C. H. N. 1939. The analysis of an animal population. J. Anim. Ecol. 8: 238—246.

Jensen, K. W. 1972. On the dynamics of an exploited population of brown trout *Salmo trutta* L. Rep. Inst. Freshw. Res., Drottningholm 52: 74—84.

— 1977. On the dynamics of the population of brown trout, *Salmo trutta* L., in Lake Øvre Heimdalsvatn, Southern Norway. Rep. Inst. Freshw. Res., Drottningholm, 56: 18—69.

— and E. Snekvik. 1972. Low pH levels wipe out salmon and trout populations in southern Norway. Ambio 1: 223—225.

Jonsson, B. 1976. Comparison of scales and otoliths for age determination in brown trout *Salmo trutta* L. Norw. J. Zool. 24: 295—301.

Leivestad, H., G. Hendrey, I. P. Muniz and E. Snekvik. 1976. Effects of acid precipitation on freshwater organisms. p. 87—111. *In* Impact of acid precipitation on forest and fresh-water ecosystems in Norway. Ed.: F. H. Brække. SNSF-project Res. Rep. 6/76. Oslo—Ås.

Mortensen, E. 1977. Density-dependent mortality of trout fry (*Salmo trutta* L.) and its relationship to the management of small streams. J. Fish Biol. 11: 613—617.

Nordeng, H. 1961. On the biology of char (*Salmo alpinus* L.) in Salangen, North Norway. I. Age and spawning frequency determined from scales and otoliths. Nytt. Mag. Zool. 10: 67—123.

Ricker, W. E. 1975. Computation and interpretation of biological statistics of fish population. Bull. Fish. Res. Bd Can. 191. 382 p.

— 1979. Growth rates and models. p. 677—743. *In* Fish Physiology. Vol VIII. Bioenergetics and growth. Eds.: W. Hoar, D. S. Randall and J. R. Brett. Academic Press, New York.

Rosseland, B. O., I. H. Sevaldrud, D. Svalastog and I. P. Muniz. 1980. Studies on freshwater fish populations — Effects on reproduction, population structure, growth and food selection. p. 336—337. *In* Ecological impact of acid precipitation. Proc. Inst. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

RYAN, P. M. and H. H. HARVEY. 1980. Growth responses of yellow perch, *Perca flavescens* (MITCHILL), to lake acidification in the La Cloche Mountain Lakes of Ontario. Env. Biol. Fish. 5: 97—108.

SEVALDRUD, I. H. and I. P. MUNIZ. 1980. Acid lakes and the inland fisheries in Norway. Results from interview studies 1974—79. SNSF-project, IR 77/80. 92 p. (In Norwegian with English summary.)

SNEKVIK, E. 1974. Acid lakes and fish stocks in Rogaland, Vest-Agder, Aust-Agder, Telemark. Compilation of information obtained from the inland fishery boards in the four counties. Report Directorate for Wildlife and Freshwater Fish, Ås. 50 p. (Mimeographed in Norwegian.)

WRIGHT, R. F. and O. K. SKOGHEIM. 1983. Aluminium speciation at the interface of an acid stream and a limed lake. Vatten 39: 301—304.

Liming of a Small Acidified River (River Anråseån) in Southwestern Sweden, Promoting Successful Reproduction of Sea Trout (*Salmo trutta* L.)

B. INGVAR ANDERSSON, INGEMAR ALENÄS and HANS HULTBERG

Swedish Environmental Research Institute, P. O. Box 5207, S-402 24 Gothenburg, Sweden

ABSTRACT

The acidification severely disturbed natural reproduction of sea trout (*Salmo trutta* L.) in the River Anråseån during the 1970's. The population decline was reversed by extensive lime applications during 1978—82.

The liming strategy included application to most headwater lakes whereby improvements of pH and alkalinity were achieved in the lower stream sections where the important reproduction occurred. Stream stretches that were highly productive increased by five times and the total length of stream stretches with reproduction doubled. Not only the total production increased but also the productivity at specific stations. This was most notable at one station in the tributary River Lerån. Densities of about 20 1+ fry per 100 m² before liming increased to approximately 40 per 100 m² the first year following liming. The density thereafter remained at this level throughout the study. Acid events during autumn and spring limited the anticipated duration of favourable water chemistry. Consequently, decreased smolt production occurred in the upper spawning areas, which, however, were less important to the total production of the river.

The overall positive effects of the limings were a significant increase of the smolt output and a more successive increase of the migrating spawners.

I. INTRODUCTION

Adverse effects of the ongoing acidification of certain surface waters are generally publicized, including such effects as damage to fish populations (HULTBERG and STENSON 1970; BEAMISH and HARVEY 1972; ALMER *et al.* 1974; MUNIZ and LEIVESTAD 1980). For streams, a decrease in reproductive success and a progressive decline in populations of Atlantic salmon (*Salmo salar* L.), sea trout (*Salmo trutta* L.), and brown trout (*Salmo trutta* L.) have been observed. JENSEN and SNEKVIK (1972) reported a drastic decline in the Atlantic salmon fishery in southern Norwegian rivers. A number of these large rivers have virtually lost their reproducing populations of Atlantic salmon. The same observations have been made in Nova Scotia (HARVEY 1980) and Sweden (EDMAN and FLEISCHER 1980) and problems are emerging also in some Scottish salmon rivers.

On the Swedish west coast, especially in the county of Bohuslän, a number of small rivers and streams are of utmost importance for sea trout production. This is also a region where acid loading has been consistently high since at least the early 1960's. Declining populations and reproductive failures of sea trout have been observed since about 1970, apparently due to acidification of the spawning streams in combination with overfishing of the stocks.

River Anråseån, which is classified as one of the most important streams in Sweden for sea trout production, has been subject to progressive acidification (HULTBERG 1984). Effects on reproduction were already observed in the years 1969 to 1971 (HULTBERG unpubl. data, cf. HULTBERG 1977). A few years later, the decline became further apparent when assessing the numbers of both spawning adults and sea-running smolts in a trap close to the river mouth. Annual migration of spawners decreased successively from about 1500 to about 700 during the 1970's.

The declining smolt production was even more pronounced. The normal number recorded between 1965 and 1976 was 1500 to 2000, whereas in 1969, 1970 and 1974 only 500—200 were registered. In 1977 and 1978 the numbers were 37 and 0, respectively. The apparent cause for this is acidification. The river water was very acid during the winter 1977 and 1978 and the draught periods in 1975 and 1976 have also contributed.

In order to prevent the complete eradication of the sea trout population in River Anråseån, anglers initiated a liming program with the objective to counteract the acidification and restore the high natural production of the stream. Funds were made available in 1977 by the Swedish government liming program.

II. THE RIVER ANRÅSEÅN WATERSHED

River Anråseån is a typical small Swedish west coast stream about 50 km north of Gothenburg. The watershed covers 103 km² on the western slopes of a highland area reaching an altitude of 170 m above sea level. About 5 km upstream of the mouth, the river divides into three major tributaries (Fig. 1). The most important spawning beds and production areas are those upstream. The flow through pronounced valleys, leading to a coastal flat west of their confluence. About 50 small (0.1—65 hectares) oligotrophic forest lakes are a significant feature of the watershed.

Geology and vegetation

The bedrock of the area is dominated by two major gothnian rock series, traversing the area in a north-south direction. Dominants are granitic and gneissic rocks but intrusions of *e.g.* granodiorite also occur. In the highlands the topography is in general small-scale but very steep. Rock outcrops are a prominent feature, as are small bogs and fens in the depressions. The till, mostly of local origin, is generally thin but deeper glacial or glacio-fluvial deposits are found, mostly at the slopes of the valleys. The bottom substrata of the tributary valleys are partly deposits of marine clay (most prominent within the valley of the tributary Lerån, where consequently agriculture is now dominating). The dominant soil types within the headwater areas are humic or ironhumic podsols.

The vegetation is a mixed coniferous forest, ranging from dry to wet associations, with either Scots pine (*Pinus silvestris* L.) or Norway spruce (*Picea abies* L. Karst.) as dominant species. The deciduous fraction is variable and mostly dominated by birch (*Betula pubescens* Ehrh. and *B. verrucosa* Ehrh.) Along the streams, however, alder (*Alnus glutinosa* L. Gaertn.) is abundant. Stands of oak (*Quercus robur* L.) are an increasingly conspicuous component in the lower coastal area.

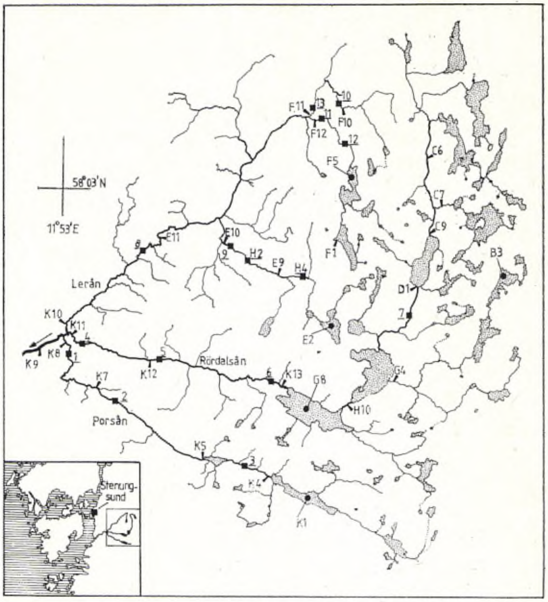


Fig. 1. Map of the upper part of the River Anråseån. The inserted map shows position at Hakefjorden. Arrows denote water chemistry stations and squares electrofishing stations.

Hydrology

Precipitation in this area varies from approximately 800 mm · year^{-1} on the coastal flat to about 950 mm (Hultberg and Andersson 1982) in the highlands. Yearly mean temperature is about 6.6 °C in the highlands and somewhat higher in the near-coast area. This difference is most notable during the winter as more snow accumulates and lasts longer in the headwater areas.

The long-term value for runoff is estimated to be 16—17 $l \cdot s^{-1} \cdot km^{-2}$ in these headwater areas (Hultberg and Andersson 1982) but is substantially less in the low altitude areas. However, during the project period 1978—82 annual precipitation has been unusually high. Thus annual runoff has been in the range 20—22 $l \cdot s^{-1} \cdot km^{-2}$ in the headwater areas (Johansson pers comm).

Much of the precipitation falls during late summer and autumn. The successive refilling of groundwater and surface reservoirs generally results in a pronounced peak runoff in late autumn (November). Depending on temperature conditions during winter, snow accumulation and snowthaw floods may be rather variable. For example, during the winter of 1976/77, the snow equivalent of about 200 mm of water accumulated and the spring thaw proceeded rapidly. During "normal" winters less snow accumulation occurs (20—40 mm as water). Occasionally, several accumulation and thaw periods may occur during the same winter.

III. MONITORING PROGRAMS

The stations selected for monitoring water chemistry and electrofishing were the same as used in an earlier (1969—72) study (HULTBERG unpubl.; HULTBERG 1977).

Water chemistry

The basic water chemistry program for the streams included 22 stations (Fig. 1). The following were analysed six times per year and most frequently during the spring: pH, alkalinity, specific conductivity, colour, Ca and Mg. Methods used were generally according to Swedish Standards except for colour which was measured spectrophotometrically at 400 nm and by a standard curve transferred to mg Pt $\cdot$ l^{-1}.

In addition, one reference (acid) lake and four limed lakes were monitored during both the turnover and stratified periods. In these lakes, oxygen, pH and alkalinity were measured along depth profiles. At the autumn turnover the regular program was expanded with analyses of: Cl, SO_4, total-P, total-N, NO_3-N, Na and K.

Troughout the period interested anglers were actively engaged in the project. They organized an annual autumn water chemistry collection comprising some 95 stations covering the entire watershed. Parameters analysed were the same as for the basic program. Furthermore, they performed electrofishings in the lower section of River Anråseån to decimate predatory northern pike (*Esox lucius* L.) prior to the sea-run of smolts.

Electrofishing

Fifteen stations were surveyed yearly, in general during September. The distribution of these stations among the three tributaries is shown in Fig. 1. In River Lerån (the most important stream for sea-trout production) 7 of the 8 stations were located higher up in second order streams. Characteristics of the stations are given in Table 1.

Fishing was performed by using a standard electrofishing gear (500 V continuous DC current). Stretches of 35 to 50 m were closed with nets and the population estimated by the removal method (cf. ZIPPIN 1956; BOHLIN 1981). Usually three consecutive fishing trials were performed. The fish caught at each effort were identified, measured, and finally returned within the same stretch. Age classification was based on size class (10 mm intervals) frequencies. The error introduced using this method was negligible as overlap between year classes was generally insignificant.

IV. LIME TREATMENTS

The initial 1978 liming covered the head waters down to the confluence of the tributaries. For the acid lakes, a dose of 12..5 g $\cdot$ m^{-3} as $CaCO_3$ was used. The persistent acid-loading was estimated to require an annual dose of 50 kg $\cdot$ ha^{-1} for neutralization.

The strategy was aimed at applying lime to most of the 50 lakes. This was based on the assumption that these lakes would act as limed reservoirs and their runoff would provide enough alkalinity for neutralization downstream. In case of lakes with short residence time, liming was carried out in lakes further upstream.

Liming commenced in August 1978 and was completed in February 1979. The total amount, 1809 tons, included about 1460 tons in the lakes and 350 tons in streams and surrounding fields (Fig. 2). A large part of the lime application was by helicopter. A standard product, $CaCO_3$ 0—0.5 mm (grain size), was used.

The estimated period of non-acid conditions was at first 5 years, but later estimates of acid loading revealed a higher dose requirement than earlier expected. The monitoring also showed this clearly and repeated liming treatments were necessary.

Table 1. *Characteristics of the electrofishing stations in River Anråseån.*

Station	Stream	Length (m)	Area (m^2)	Mean depth (m)	Water velocity characteristics	Vegetation and bottom character
1	Porsån	40	140	0.35	Mainly swift current, minor rapids occur	Sheltering trees. Large cobbles, minor sand-clay areas
2	,,	50	150	0.15	Slow to swift current	Sheltering trees and bushes. Mainly sand-gravel beds
3	,,	50	75	0.10	Mostly rapids with torrent current	Sheltering bushes. Large cobbles, some bryophytes
4	Rördalsån	41	220	0.30	Swift, and partly rapid current	Sheltering trees. Sand-gravel and larger cobbles
5	,,	39.5	225	0.40	Rapids with partly torrent current	Sheltering coniferous trees. Large cobbles, some bryophytes
6	,,	50	100	0.25	Swift current	Partly tree-sheltered. Cobbles, sand, and clay
7	"Nordån"	50	100	0.15	Slow to swift current	Sheltering trees. Cobbles and sand, some bryophytes
8	Lerån	35	215	0.40	Slow to rapid current deeper pools	Partly sheltered by trees. Cobbles with sand-clay spots
9	Labolbäcken	36	105	0.25	Slow to rapid current	Sheltering bushes. Smaller cobbles, sand and clay
H2	,,	50	128	0.20	Slow to rapid current	Sheltering trees. Smaller cobbles, gravel with sand
H4	,,	50	100	0.15	Swift current	Tree- and bush-sheltered. Mainly sand, smaller cobbles
10	Abborrtjärnsbäcken	40	84	0.15	Swift to torrent current	Sheltering trees and bushes. Cobbles to sand
13	,,	50	100	0.30	Slow, with deeper pools	Bushes. Clay with small sand-gravel areas
11	Kvarnsjöbäcken	40	100	0.30	Slow, with deeper pools	Partly sheltered. Mainly clay, minor spots with sand, small cobbles
12	,,	48	95	0.15	Slow to rapid current	Bush and tree sheltered. Smaller cobbles sandy

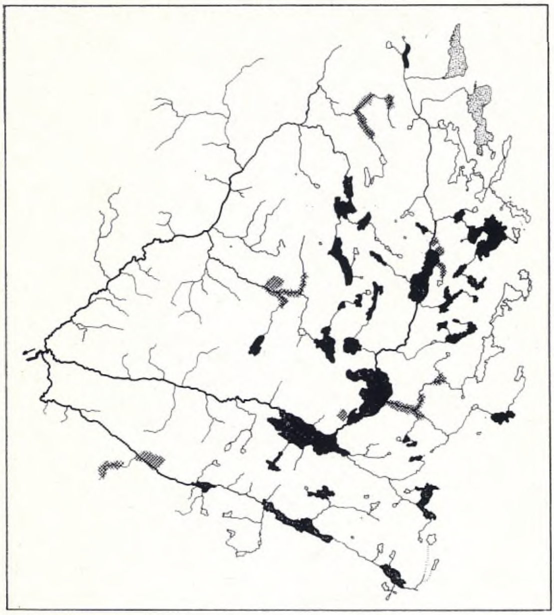


Fig. 2. Lime applications 1978/79 in lakes (black) and streams (cross-hatched). Stippled lakes were limed 1974 for a research project (Hultberg and Andersson 1982).

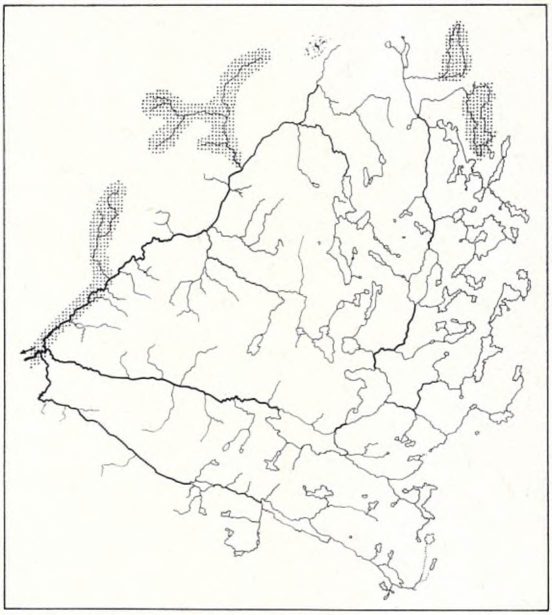


Fig. 3. Areas of River Anråseån having pH-values > 6.0 in November 1977.

Complementary applications were performed annually in Lake Lillevatten, and in 1980 and 1981 in Lakes Håltesjön and Bredvatten, respectively. Finally, a repeat liming was performed in the spring of 1982, when 722 tons were applied as in the earlier application, though the product was finer (grain size 0—0.2 mm).

V. LIMING EFFECTS ON WATER CHEMISTRY

Before the liming of River Anråseån in 1977 virtually all surface waters of the watershed had pH-values less than 6.0 (Fig. 3), with some values being as low as 4—4.5 in headwaters. The only exceptions were some lakes where liming activity had already begun (Lakes Bredvatten and Lysevatten) (Hultberg and Andersson 1982).

By November 1978, most of the first lime application was complete and the pH was greater than 6.0 in almost all of the reproduction areas downstream of the limed lakes. Exceptions were the streams Abborrtjärnsbäcken (F10) which was not yet treated and Labolbäcken (E9) where the influence of the liming in the upstream lake had not yet taken effect (Fig. 4). In this upstream lake (Valevatten E2), a large fraction of the lime settled to deep water resulting in a comparatively slow but progressive dissolution (Fig. 5). In addition, this delayed response was even more pronounced since a small acid lake downstream Lake Valevatten was not treated.

Liming induced an almost instant increase in lake and stream water pH and alkalinity as has been generally observed elsewhere (National Swedish Board of Fisheries and National Swedish Environmental Protection Board 1981; Hultberg and Andersson 1982). Alkalinity levels were in the range 100—300 mikroequivalents · L^{-1} for the limed lakes, whereas the downstream effects were variable depending on specific conditions. Inflow from unlimed brooks and acid seepage water decreased pH and alkalinity. In some downstream stretches, however, the baseflow from marine clay deposit areas substantially increased the alkalinity (*e.g.* along River Lerån).

During the period following the lime treatments, alkalinity levels decreased progressively, due to continuous acid loading, with water residence time as the most moderating factor. By the

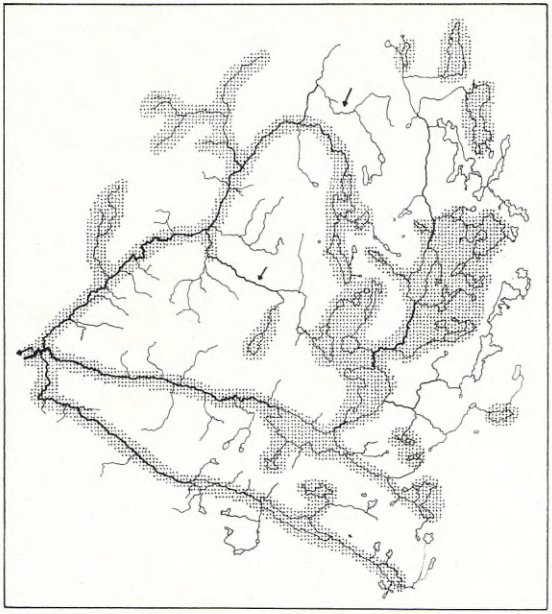


Fig. 4. Areas of River Anråseån having pH-values > 6.0 in November 1978, (after the major lime application). Arrows denote the brooks Labolbäcken (south) and Abborrtjärnsbäcken (north).

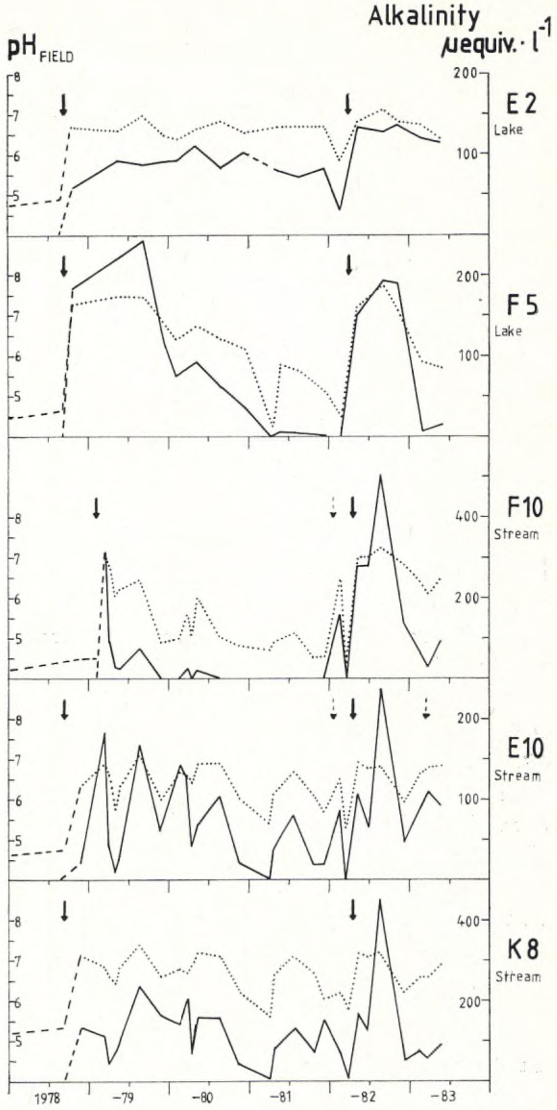


Fig. 5. Representative examples of pH (dotted line) and alkalinity (continuous line) development following the lime treatments. Bold arrows indicate direct liming of the object and broken arrows indicate upstream limings.

spring of 1981, alkalinity had decreased below the critical level required to buffer the acid snowthaw. All surficial lake waters and streams in the watershed were affected. Some were severely acidified resulting in zero alkalinity and pronounced decreases in pH *e.g.* stations F5 and E10 (Fig. 5). The entire stretch of the River Porsån was affected. Even at the lowest station (K8) the decrease in alkalinity and pH was notable (Fig. 5). The downstream stretches of Rivers Lerån and Rördalsån were, however, only slightly affected with less significant alkalinity decreases. During the spring acid event 1981, the same areas that had pH-values less than 6.0 prior to liming, had returned to their original state. In general, the pH of the lakes and streams increased during summer, but in some lakes, *e.g.* Lake Kvarnsjön, alkalinity remained zero (F5 in Fig. 5).

Heavy rains further reduced the general pH-level of the river during the autumn of 1981. As can be seen from Fig. 6 most headwaters had pH less than 6.0 by November. Exceptions were Lakes Valevatten and Långevatten which have comparatively long residence times and the newly re-limed Lake Bredvatten. The lower and most important reproductive stretches of Rivers Lerån and Rördalsån were also less affected. River Porsån had a pH less than 6.0 throughout its range.

The response to re-liming during spring 1982 was in general similar to that during the first liming even though smaller amounts were applied. Clearly, this was somewhat dependent on the

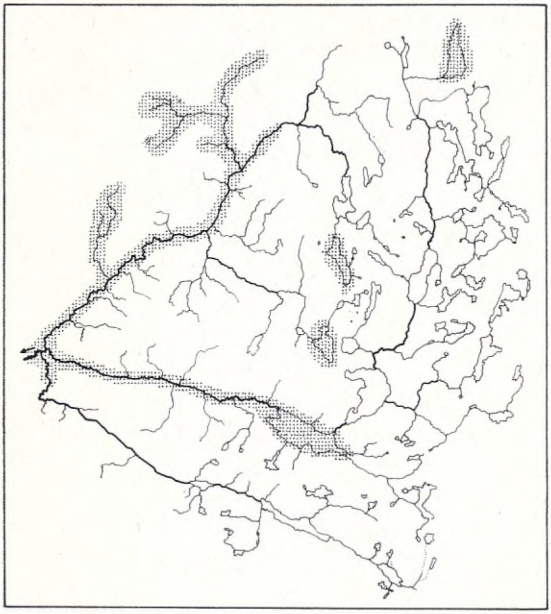


Fig. 6. Areas of River Anråseån having pH-values > 6.0 in November 1981.

remaining effects from the first application in 1978/79.

In case of the small headwater brook Abborrtjärnsbäcken the application procedure was altered. The main fraction of lime was spread in the upper stretches, where the brook seeps through a vast fen area. The same amount of lime (80 tons) resulted in an improved effect which may turn out to be more permanent.

VI. LIMING EFFECTS ON SEA TROUT REPRODUCTION

Results from electrofishing at the 15 stations are summarized in Table 2. In order to better understand the changes observed a few typical cases will be dealt with in more detail.

Station 1 in River Porsån is situated close to the confluence of the tributaries. In 1977, before the lime application, a single young fish (probably 1+ or 2+) was caught. Apparently, despite low pH, the sea-trout spawning has continued within this stretch of stream.

Normally, hatching and growth in the first summer may be possible during moderate acidification. The acid events, however, during the winters would have decimated one-summer-old fry populations each year. Hence, before liming, only small numbers of older sea-trout fry may be observed together with O⁺ fry. Increased acidification then would result in decreasing numbers of O⁺ fry and few or no 1+ fish as was the case at station 1 in 1977.

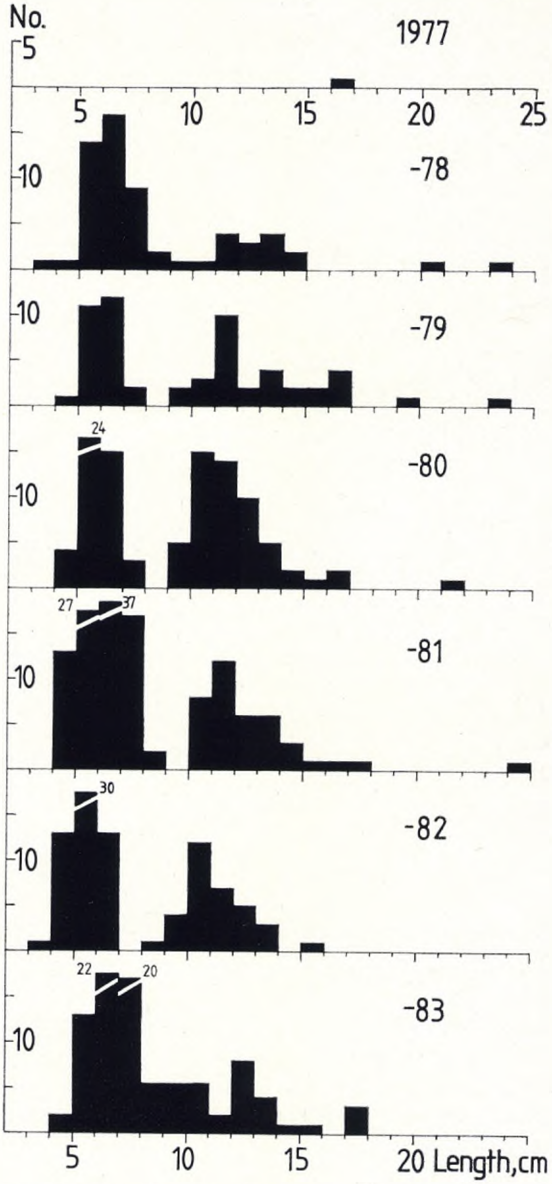


Fig. 7. Size frequency distribution of sea trout (*Salmo trutta* L.) fry at station I during 1977 to 1983.

Table 2. *Density (number per 100 m^2) of sea-trout fry at the electrofishing stations during 1977—83.* The density assessments are calculated according to Zippin (1956).

Station	Category	1977	1978	1979	1980	1981	1982	1983[1]
1	0+	—	33	19	36	77	51	61
	total	—, 1	45	45	74	101	69	75
2	0+	—	—	6	5	17	17	—
	total	—	—	17	8	24	29	—
3	0+	—	—	1 fry	—	—	—	N.F.
	total	—	—	—	—	—	—	N.F.
4	0+	17, 12	25	43	4[2]	112	54	53
	total	34, 19	44	63	15	121	71	61
5	0+	1, 9	8	14	5[2]	28	17	> 8
	total	14, 20	15	22	8	32	31	23
6	0+	—	—	9	7	12	45	N.F.
	total	1 fry	—	15	8	12	46	N.F.
7	0+	—	—	5	—	8	6	N.F.
	total	—	—	6	3	8	8	N.F.
8	0+	N.F.	34	49	27	61	128	53
	total	N.F.	54	74	60	93	166	93
9	0+	—	11	90	52	56	87	5 †
	total	3	11	102	85	91	117	48
H2	0+	—	—	135	—	53	13	45
	total	—	—	153	65	65	22	55
H4	0+	—	—	26	N.F.	78	4	N.F.
	total	—	—	26	N.F.	80	29	N.F.
10	0+	—	—	31	10	—	1	163
	total	—	—	42	27	6	9	166
13	0+	N.F.	N.F.	12 ††	32	99	69	75
	total	N.F.	N.F.	28	54	143	115	146
11	0+	—	—	21	23	109	39	66
	total	—	—	46	41	126	80	99
12	0+	—	—	18	—	34	1	N.F.
	total	—	—	26	17	39	14	N.F.
Annual mean	0+	1.2	7.4	31.9 ± 9.4	14.4 ± 4.3	49.6 ± 10.1	35.5 ± 9.7	58.8 ± 15.3
± S.E.	total	3.5	11.2	44.3 ± 10.4	33.2 ± 7.6	62.7 ± 12.5	53.7 ± 13.9	85.1 ± 15.5

[1] Fishing in late October
[2] Fishing at highwater
† Probably influenced by present spawners
†† Based on only one fishing occasion

N.F. = no fishing performed, — catch was zero
Two fishings were performed at stations 4 and 5 in 1977

The improved water chemistry at station 1 after liming altered the situation significantly. The total catch in 1978 was 45 per 100 m^2. This included 1+ fry that, considering the observations in autumn 1977, apparently must have reached the stream stretch at station 1 by migration (Fig. 7). This stretch was almost nonproductive before liming but successively improved thereafter. The annual densities of 1+ fry varied between 15—40 per 100 m^2 during 1979—83.

Station 11 in a smaller brook, Kvarnsjöbäcken, higher up along River Lerån, was severely acidified around 1970 (Hultberg, unpubl.). Densities of sea-trout fry already then were low and later no fry survived in these upper reaches. After liming no spawning had occurred in 1978, so in May

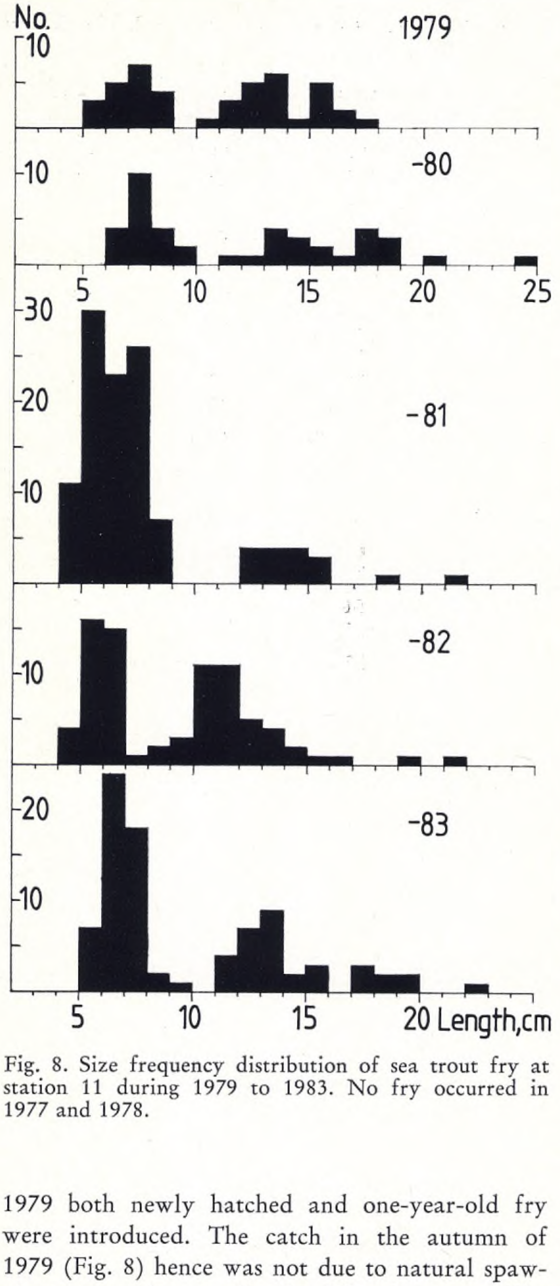


Fig. 8. Size frequency distribution of sea trout fry at station 11 during 1979 to 1983. No fry occurred in 1977 and 1978.

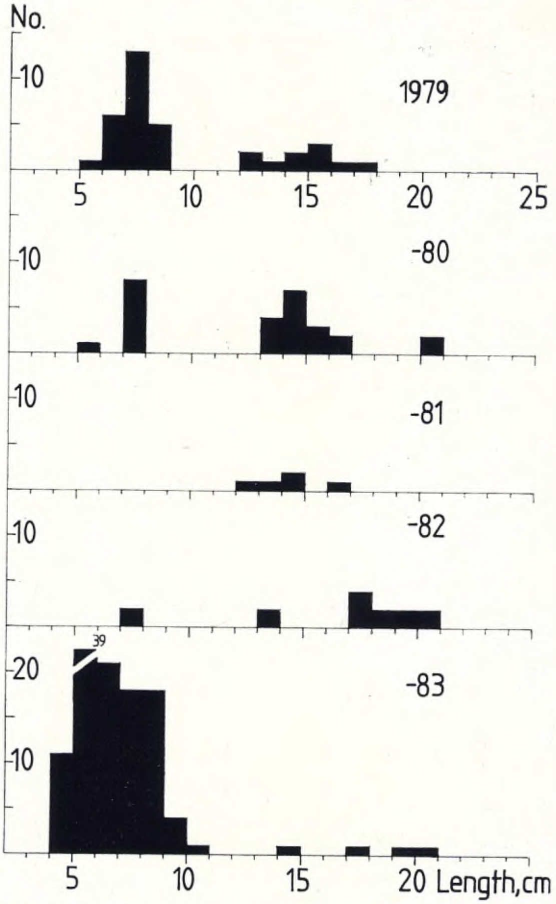


Fig. 9. Size frequency distribution of sea trout fry at station 10 during 1979 to 1983. No fry occurred in 1977 and 1978. Note successive decline and significant increase in 1983 after repeated liming upstream.

1979 both newly hatched and one-year-old fry were introduced. The catch in the autumn of 1979 (Fig. 8) hence was not due to natural spawning as was the case for the following years. The year 1981 was an exceptionally good year with a density of 109 0+ fry per 100 m². Densities of the naturally reproduced 1+ fry increased from about 20 to about 40 per 100 m² between 1981 and 1983. Apparently sea-trout reproduction at this lowest station was only to a minor degree affected by the re-acidification of Kvarnsjöbäcken.

At the upstream station (12), reproductive success was clearly more affected, as the density decreased to a third (Table 2).

Prior to liming, reproduction at station 10 was similar to that at station 11. Also the response immediately following liming was similar with no natural spawning. Stockings in the spring of 1979 resulted in comparatively low densities, 31 0+ and 11 1+ per 100 m² (Fig. 9). The liming effect in the Abborrtjärnsbäcken brook was found to be diminishing already by 1979 (cf Fig. 5, Stn, F10). Consequently, natural reproduction was essentially unsuccessful during 1980 to 1982, with total densities of 27, 6 and 9 fry per 100 m². The second liming in 1982, however, resulted in

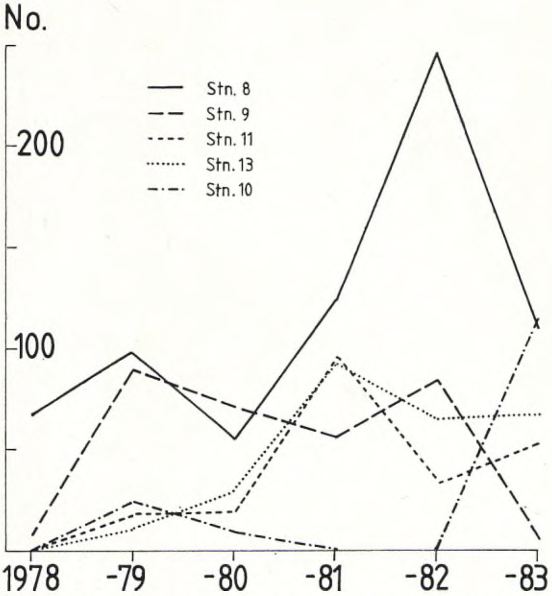


Fig. 10. Number of 0+ sea-trout fry caught at different electrofishing stations in River Lerån during 1978 to 1983.

a very successful hatching in 1983 with a 0+ density of 163 per 100 m². In 1982, a number of larger (170—210 mm) fishes (about 2+ or more years old) were caught. The same was found at station 11 and apparently an increasing resident population has developed in these upper stretches.

Apart from the occasional large year to year variation of especially 0+ fry (Fig. 10) the general response to liming was positive. Among the monitoring stations, also station 6 became naturally productive again after liming.

At two other stations, H2 and H4, good or fairly good production was achieved by introduction of either fry or spawning fishes. These stations are situated above a migratory obstacle and consequently any production is dependent on management efforts. The same pertains to station 7 but here the variable pH may have hampered reproduction. It was only at station 3 where liming had no effect in the sense that no sea-trout production was observed.

VII. CONCLUSIONS

Before the liming in 1978/79, most headwaters of River Anråseån were acidified. During the winters, acid events probably affected the entire system upstream of the confluence of the tributaries and consequently sea-trout production was severely impaired. This was the result of a progressive development, that already in the early 1960's may have affected sea-trout reproduction (cf. HULTBERG 1984).

In the years after liming (1978 to 1982) the above situation was observed to be significantly changed. Improvements in water chemistry were such, that most of the important sections responsible for sea-trout production had pH-values greater than 6.0 (up to the acid events of 1981). The re-liming in 1982 again generally increased pH.

The overall effect of the liming was a substantial increase in sea-trout reproduction (observed at all stations). Interestingly production, indicated as the density of 1+ fry, also increased at the less acidified station 8 in the River Lerån.

This station is situated within the lower reaches where pH, even after heavy autumn rains, never decreased below 6.0. From a fairly good density of 20 per 100 m², a successive increase to approximately 40 per 100 m² followed after the liming (Fig. 11). It could be concluded therefore that acidification earlier had inhibited production here, even though water chemistry generally was assumed to be quite acceptable.

The production of 1+ fry after liming was approximately the same, (about 40 per 100 m²) at the stations in River Lerån at the end of the period. Liming did not improve the production in River Rördalsån at stations 4—6 in spite of much improved water chemistry (Fig. 11). In River Porsån the stretch at station 2 became productive, and at station 1 production first increased substantially, then by reacidification returned to a low level. The reasons for the observed differences between streams are not fully understood but presumably include factors such as water chemistry, food availability, habitat preferences, and interactions with a population of brown trout. Instability in spawning behaviour in the expanding sea-trout population might also contribute to this variable response as spawning migration is more easy in River Lerån than in Rivers Porsån and Rördalsån.

Combining the data for the river system as a

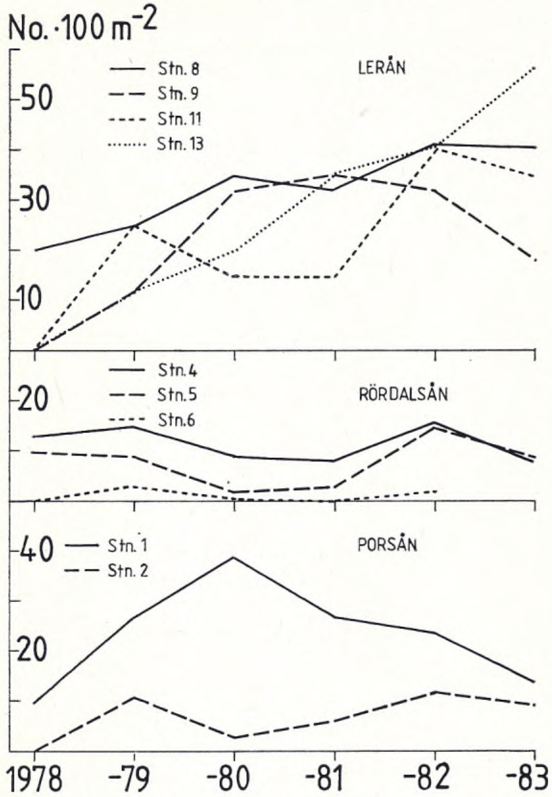


Fig. 11. Densities of 1+ sea-trout fry at selected electrofishing stations during 1978 to 1983.

whole the following can be observed: The total length of stream sections where sea-trout reproduced was approximately doubled after liming. The total stream length that was highly productive increased fivefold.

The validity of this relative to the total stock production is probably somewhat questionable, as randomized sampling was not used. This is a prerequisite for stock estimates as pointed out by Bohlin (1981).

However, as is indicated in Fig. 12, the recorded searun of smolts increased significantly after liming. This should be conclusive enough to postulate that the total production of naturally spawning sea-trout indeed increased significantly due to liming, and the stock will subsequently increase.

Experience gained thus far allow us to recommend the following: For river systems like River

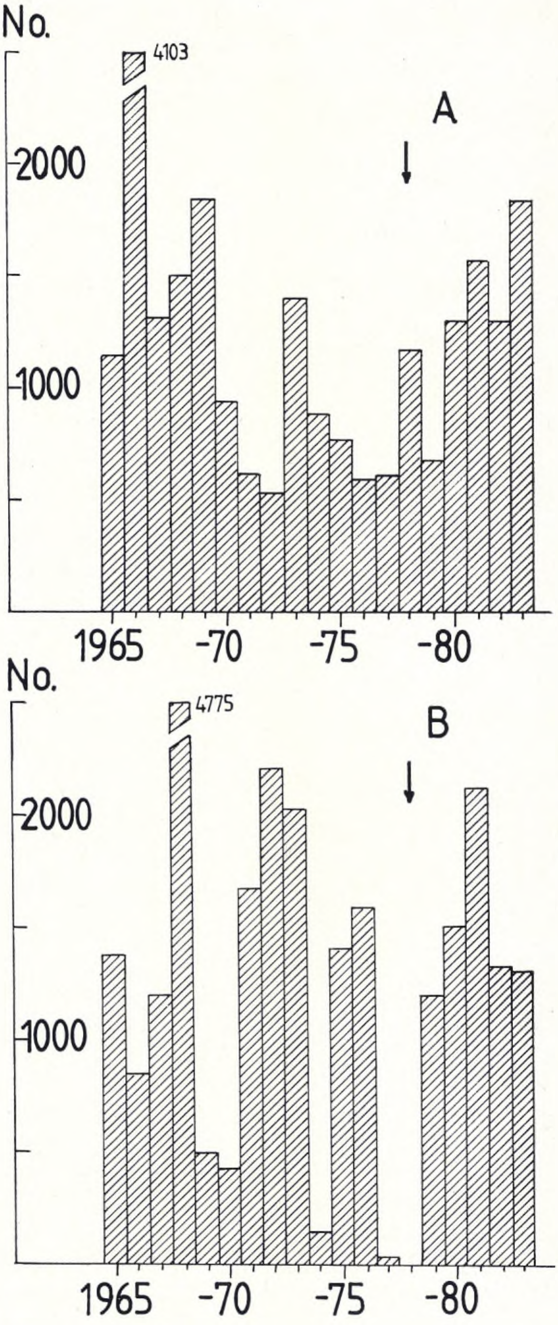


Fig. 12. Recorded number of migrating adult spawners (A) and searunning smolt (B) at a trap near the River Anråseån mouth during 1965 to 1983. Arrows indicate first liming. Note exceptional migrations 1966 (spawners) and 1968 (smolts).

Anråseån, which are located in areas with continuous high acid loading, liming should be planned for durations of only 2—3 years in order to be efficient, when lake residence times are short to moderate. The application should also be either as complete as possible, or use of costly dosing equipment high up in subsystems should be considered in order to reduce yearly acid flux (especially at snowthaw).

The main objective of the liming project in River Anråseån, which was to re-establish natural sea-trout reproduction, was achieved during this first period. Future work with continued liming will show if it is possible to achieve the probable overall maximum density of about 40 1+ fry per 100 m^2 over most of the productive stream sections. If so, River Anråseån will again rank as one of the most valuable sea-trout streams on the Swedish west-coast.

VIII. ACKNOWLEDGMENTS

We would like to express our gratitude to the STEERING GROUP of the ANRÅSE RIVER PROJECT and to all enthusiastic anglers for their devoted voluntary work. ARNO ROSEMARIN corrected the language and PER OLSÉN gave valuable comments on the manuscript, which was typed by EVA KNUDSEN and MARIE ENGHULT.

IX. REFERENCES

ALMER, B., W. DICKSON, C. EKSTRÖM, E. HÖRNSTRÖM and U. MILLER. 1974. Effects of acidification on Swedish lakes. Ambio 3: 30—36.

BEAMISH, R. J. and H. H. HARVEY. 1972. Acidification of LaCloche Mountain Lakes, Ontario, and resulting fish mortalities. J. Fish. Res. Bd Can. 29: 1131—1143.

BOHLIN, T. 1981. Methods of estimating total stock, smolt output and survival of salmonids using electrofishing. Rep. Inst. Freshw. Res., Drottningholm 59: 1—14.

EDMAN, G. and S. FLEISCHER. 1980. The River Högvadsån liming project — a presentation. p. 300—301. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

HARVEY, H. H. 1980. Widespread and diverse changes in the biota of North American lakes and rivers coincident with acidification. p. 93—98. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

HULTBERG, H. 1977. Thermally stratified acid water in late winter — a key factor inducing self-accelerating processes which increase acidification. Wat., Air, Soil Poll. 7: 279—294.

— 1984. Historical changes in fish populations and water chemistry in Lake Gårdsjön and lakes in a near area during the last century. Ecol. Bull. Stockholm 37. (In the press.)

— and B. I. Andersson. 1982. Liming of acidified lakes: Induced long-term changes. Wat., Air, Soil Poll. 18: 311—331.

— and J. STENSON. 1970. Effects of acidity on the fish fauna of two small lakes in Bohuslän, southwestern Sweden.

JENSEN, K. W. and E. SNEKVIK. 1972. Low pH levels wipe out salmon and trout populations in southernmost Norway. Ambio 1: 223—225.

MUNIZ, I. P. and H. LEIVESTAD. 1980. Acidification — effects on freshwater fish. p. 84—92. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

NATIONAL SWEDISH BOARD OF FISHERIES and NATIONAL SWEDISH ENVIRONMENTAL PROTECTION BOARD. 1981. Liming of lakes and rivers 1977—1981 in Sweden. Inform. Inst. Freshw. Res., Drottningholm (4). 201 p. (In Swedish with English summary.)

ZIPPIN, C. 1956. An evaluation of the removal method of estimating animal populations. Biometrics 12: 163—189.

The Distribution of Trout (*Salmo trutta* L.) in Relation to pH – an Inventory of Small Streams in Delsbo, Central Sweden

BO ANDERSSON[1] and PAUL ANDERSSON[2]

[1] Östra Tolbo 4662, S-820 60 Delsbo, Sweden.
[2] The National Swedish Environment Protection Board, P. O. Box 1302, S-171 25 Solna, Sweden.

ABSTRACT

In order to study the distribution of trout (*Salmo trutta* L.), 23 localities in 21 streams of a 100-km^2-large area in southern Delsbo, Sweden, were electrofished during the summer of 1978.

The trout abundance varied between 0—1.1 individuals/m^2. Few decades ago, all the streams had stocks of trout. The abundance and growth of the trout were well correlated to the water acid quality, but not with the width, depth or discharge of the locality.

There were no trout in localities with summer $pH < 6.0$ (spring $pH < 4.8$). Trout abundances were one to four times less in localities at summer $pH \leq 6.5$ (spring $pH \leq 5.1$) compared to localities of higher pH. The trout growth rate was significantly higher in localities of lower abundance. High water colour (humic substances) favoured trout growth, independent of the water acidity.

I. INTRODUCTION

Small streams constitute habitats for genetically unique strains of trout (*Salmo trutta* L.), which have become well adapted to the watercourse they live in. These strains can either be local or consist of migrating lake- and sea-living forms.

Our observations in the studied area, with many streams, showed that trout in the last decades had become progressively more patchily distributed. On the basis of these observations, it was deemed of importance to carry out further research. The purpose of this study was to determine the abundance of trout in these streams, and to look for correlations between trout abundance, trout growth rate, water chemistry (mainly pH) and physical parameters.

The research was performed in southern Delsbo, Hälsingland, situated in the east of central Sweden (Fig 1). The studied streams belong to three different catchment areas, covering an area of about 100 km^2 at 100—300 m above sea level. The bedrock is mainly granites and gneisses while the soils consist of moraine, silt and very fine sand. The three northwestern streams drain an area with sand ridges. The investigated area is forested except in some minor parts with bogs and swamps. Forestry including peat moss draining is intensive in the area.

II. SAMPLING AND ANALYSES

At 23 localities in 21 streams electrofishing was carried out by means of an electrofishing unit (make: Lugab, Ultrapuls), with pulsating direct current at 600 V, in 30—60 m sections. Prior to fishing, each section was closed by means of nets (mesh size 2 mm). The specimens from two consecutive fishings were picked up for counting and further analysis. Fishes escaped from the second fishing were included as number of individuals in the section. Sections were chosen as to provide equal areas of different water velocities. Length, weight and age of fish were determined. Otoliths were used to determine the age.

Mean stream depth and mean width (means of observations on every fifth metre) of the fished sections were measured. Stream discharge was also determined. The measurements were made by calculating the cross-sectional area and measuring the water velocity at specified sections (5—10 m) of the stream perpendicular to the cross-section (Bruce and Clark 1969).

Water was sampled at the electrofished sections at the occasion of electrofishing. In addition water was sampled at 14 of the 23 localities during May 1978. Monthly sampling of water was carried out in the streams since spring 1980. Since 1977 sporadic, and since 1980 monthly, sampling of pre-

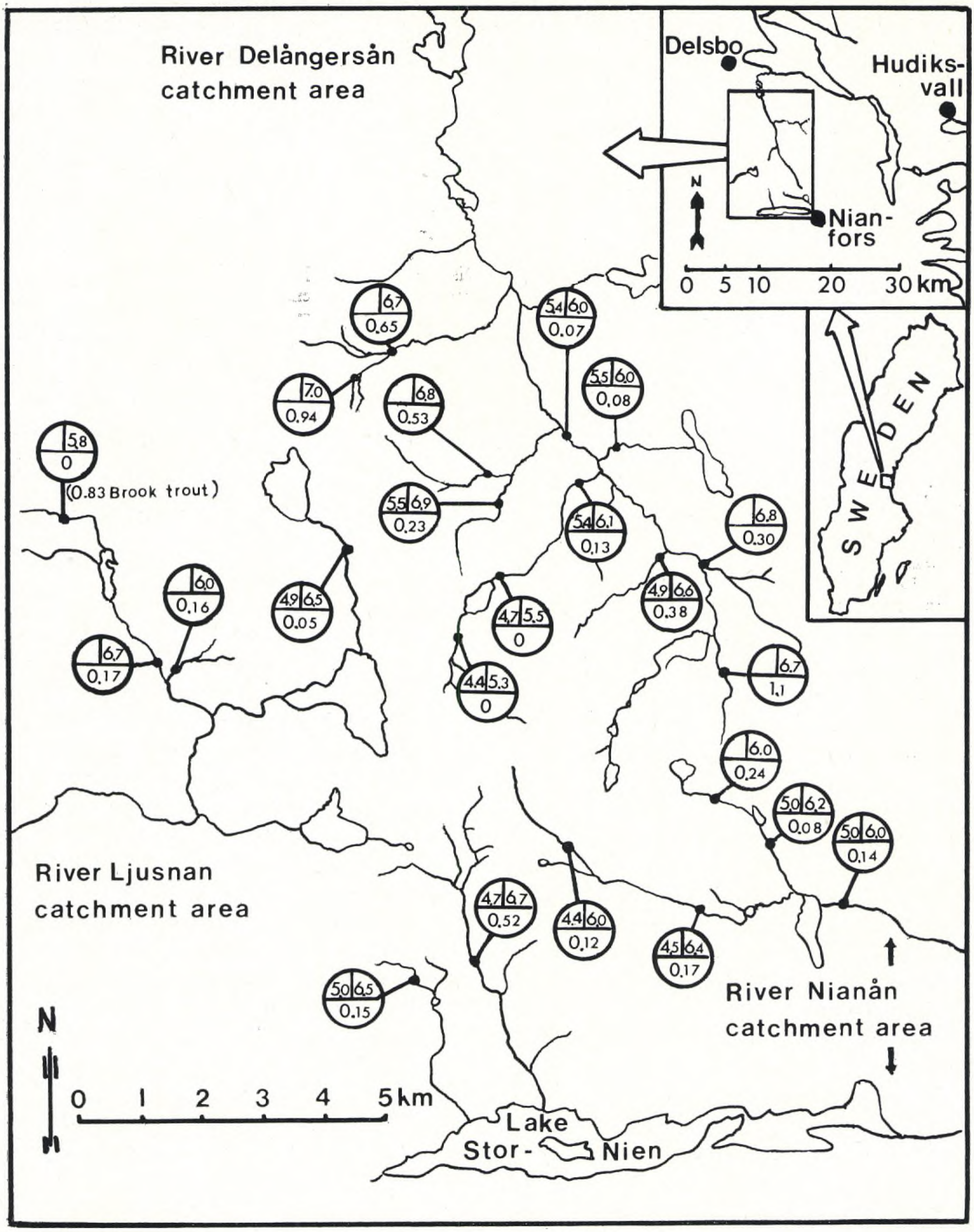


Fig. 1. Map of the studied area with trout abundances (individuals/m^2) and pH values in streams in southern Delsbo, Hälsingland.

1 = pH during spring 1978
2 = summer pH during the electrofishing period 1978
3 = trout abundance at the time of electrofishing July—September 1978

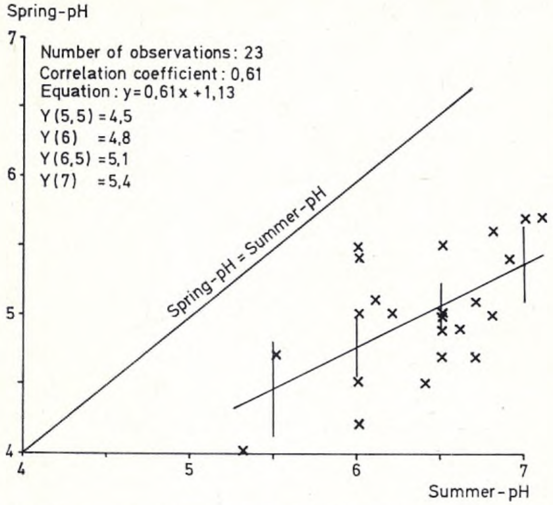


Fig. 2. Correlation between pH in summer and spring at the 23 electrofished localities. At 14 of the localities the pH values measured 1978 have been used. When spring pH data from 1978 were missing, data from lowest spring pH and highest summer pH during the year 1980 or 1981 were used in the correlation. Calculated spring pH values given at four summer pH values.

cipitation was carried out. The stream water and the precipitation were chemically analysed for pH, electro-conductivity, water colour and alkalinity according to the SWEDISH STANDARD METHODS.

The influence of pH on trout abundance and trout length is graphically illustrated. Cross-correlations have been made between trout length, trout abundance, pH, H^+ ion concentration, electro-conductivity, water colour, alkalinity, stream width, stream depth and stream discharge. Stepwise multiple regression analysis was used to interpret the relative dependence of variables studied to trout abundance and trout length. To avoid overestimation because of mathematical dependence between pH and H^+ concentration the regressions were made both with and without pH.

III. RESULTS AND DISCUSSION

Water chemistry

Fig. 1 shows that the pH value at the 23 electrofished localities varied between 5.3 and 7.0 during the summer of 1978. Spring pH measured at 14 of the localities varied between 4.4 and 5.5 during May the same year. Complementary measurements in the same streams during 1980—81 showed summer pH maxima at 7.1 and spring minima at 4.0. Apparently pH in some of the streams were lower than pH in the precipitation, which has varied between 4.2 and 4.5.

Table 1. *Mean values with standard deviations for variables correlated with trout abundances at 23 electrofished localities.*

Variable		Mean value	SD
Abundance	individuals/m^2	0.27	0.29
$H^+ \times 10^{-7}$	mol/l	8.6	11.3
Electrocond.	mS/m	3.0	0.5
Colour	mg Pt/l	150	70
Alkalinity	meqv HCO_3^-/l	0.06	0.05
Discharge	l/s	60	50
Stream width	cm	190	100
Stream depth	cm	22	9
pH (median value)		6.5	—

In order to illustrate the fluctuations of pH in the area during the year, a plot of spring pH versus summer pH has been constructed (Fig. 2). The greatest fluctuations were observed in streams with high summer pH values. However, the relative fluctuation in H^+ ion concentration was greater in the streams of low summer pH.

The alkalinity in the streams during the summer of 1978 was between < 0.01 and 0.16 meqv HCO_3^-/l. The electro-conductivity ranged between 2.2 and 4.0 mS/m and the water colour between 90 and 240 mg Pt/l during the same period (Table 1), which showed that the streams were relatively poor in dissolved salts and rich in humic substances.

Trout abundance

The trout abundance was between 0 and 1.1 individuals/m^2 (Fig. 1). The abundance was comparable with that in streams in central and western Sweden (BERGQUIST pers. comm.) and with three streams on the Swedish West Coast before liming, but lower than after liming (THÖRNELÖF, unpubl. data).

All the streams in the study have had trout. At two of the three localities missing trout in 1978, trout were caught in 1970 and the year before the research project started, respectively. The third

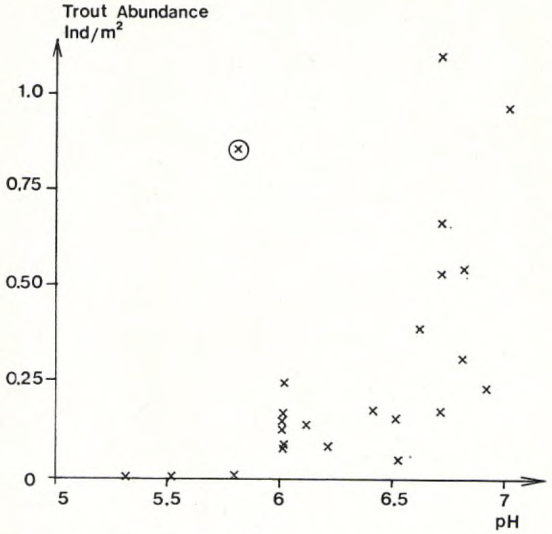


Fig. 3. The abundance of trout at different pH values during the summer of 1978 in streams in southern Delsbo.
⊗=brook trout (*Salvelinus fontinalis*).

locality with no trout was one of those most rich in trout before peat moss draining was carried out in the precipitation area in the 1940s. Observations of the trout abundance made by ourselves and our friends when fishing in the streams during the last 25 years, and even older observations made by other local residents (pers. comm.), clearly indicated that the trout abundance had been reduced in most streams where trout were still present.

Trout abundance and pH

Fig. 3 shows the trout abundance plotted against pH. The three localities with no trout had a summer pH value below 6.0. The localities with summer pH > 6.5 were about one to four times richer in trout than those with summer pH 6.0—6.5.

pH values of 5 or lower may cause mortality to eggs and fry of trout (LEIVESTAD *et al.* 1976, WRIGHT and SNEKVIK 1970). The hatching of the eggs occur during spring when pH is at its lowest. On the basis of the relationship shown in Fig. 2, the summer pH values were transferred to approximate spring pH values. The lack of trout at localities with spring pH < 4.8 and low observed abundance at spring pH 4.8—5.1, might be explained by failed or limited reproduction in the streams.

Streams which are easily recolonized from lakes and tributaries with more stable pH conditions had more trout than could be expected despite their low pH values during spring. Other explanations to deviations from the proposed restricting pH value of approximately 4.8 might be differences in acid tolerance of the trout strains, or differences in the humic content and/or aluminium concentrations in the streams.

No aluminium was analysed in this study, but later investigations in some of the streams, May 1982, have shown high aluminium concentrations (100—380 µg/l), but with no correlation between pH and aluminium concentration. The humic content was rather high (colour: 40—160 mg Pt/l) indicating aluminium to be organically complexed to a considerable degree. However, when compared to values from South-Swedish lakes, for which aluminium speciation has been carried out (BJÄRNBORG pers. comm.) the aluminium concentrations were comparatively higher than would be expected from the colour values.

Toxic actions by aluminium and also iron suspensions, which damage gill mucus and filaments, were found in natural waters at changing and moderately low pH values (ALMER *et al.* 1978, MUNIZ and LEIVESTAD 1979, SYKORA *et al.* 1975, DICKSON 1979, DRISCOLL *et al.* 1980).

It should also be mentioned that one of the streams from which trout have disappeared had a strain of brook trout (*Salvelinus fontinalis* MITCHILL) which is known to be less sensitive to acid water than brown trout (LEIVESTAD *et al.* 1976, MUNIZ and LEIVESTAD 1979).

Correlations between trout abundance, water chemistry and environmental variables

Table 1 shows the mean values and standard deviations for the variables studied. The dependence of trout abundance on stream acidity was confirmed by good correlations ($p < 0.025$) between trout abundance and pH ($r=0.67$), H^+ ion concentration $r=-0.46$) and alkalinity ($r=0.64$), respectively. In spite of good correlations between electro-conductivity and the chemistry variables mentioned above, no direct correlation was obtained between electro-conductivity and trout abundance. The stepwise multiple regressions in-

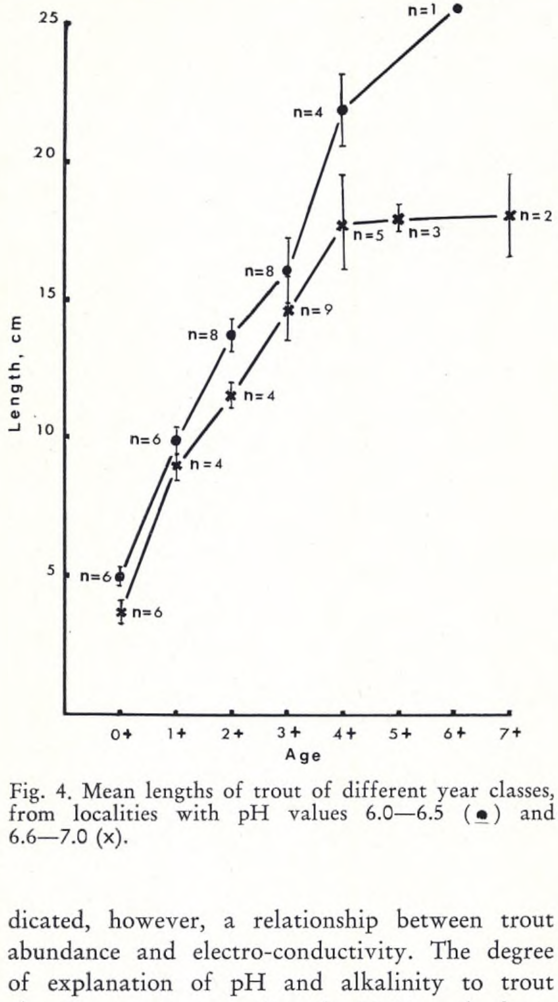


Fig. 4. Mean lengths of trout of different year classes, from localities with pH values 6.0—6.5 (●) and 6.6—7.0 (x).

dicated, however, a relationship between trout abundance and electro-conductivity. The degree of explanation of pH and alkalinity to trout abundance was approximately 50 % and of another 5 % when electro-conductivity was included. The remaining variables only increased the degree of explanation by a further 6 %. The correlations with stream width and discharge, although weak, showed that trout abundance was favoured by narrow streams with low discharge. Water colour and stream depth were not correlated to trout abundance.

This relationship between trout abundance and low electro-conductivity at low pH values can be explained as an inhibition by the H^+ ion, of the active uptake of sodium through the gills. The inhibitions together with the low ionic strength of the water leads to a decrease in the sodium and chloride content of the blood, causing loss of salts from the whole fish body (LEIVESTAD *et al.* 1976, McWILLIAMS 1980a). Both field studies and laboratory experiments show that fish have a better resistance to acid exposure in the presence of calcium (WRIGHT and SNEKVIK 1978, McWILIAMS and POTTS 1978, McDONALD *et al.* 1980), which might also be an effect of increased ionic strength. Adaption of trout to acid water is found to some extent and depends on how quickly the changes in pH occur and on changes in gill permeability to sodium (McWILLIAMS 1980b).

Trout growth and correlations to water chemistry and environmental variables

The length of trout of the same age varied between the localities. Fig. 4 shows that the mean lengths of trout at localities in the pH interval 6.0—6.5, where trout reproduction was probably disturbed, were greater than in the pH interval 6.6—7.0. This observation was confirmed by significant ($p < 0.05$) correlations between the length of four-summer-old (3+) trout and the abundance ($r = -0.48$) and the abundance-restricting water chemistry variables pH ($r = -0.53$), H^+ ion concentration ($r = 0.59$), alkalinity ($r = -0.52$) and electro-conductivity ($r = 0.50$). The correlations also showed that high water colour in spite of no correlation with the abundance-restricting variables favoured trout growth ($r = 0.52$). The stepwise multiple regressions explained the variations in trout length to 32 % by H^+ ion concentration. The degree of explanation increased to 51 % when also the water colour was considered. Totally about 70 % of the variation in trout length could be explained by the variables studied. The trout length was not correlated with width, depth or discharge of the stream.

Increased growth of fish as a consequence of disturbed reproduction and decreased competition for food in sparse populations at decreasing pH, buffering capacity and ionic strength was also observed in Scandinavian lakes as a first symptom of acidification (JENSEN and SNEKVIK 1972, ALMER *et al.* 1978). Reduced growth in spite of good supply of food was observed in acid lakes in Norway and Canada (ROSSELAND 1980, ROSSELAND *et al.* 1980, BEAMISH *et al.* 1975) and at acid exposure in laboratory experiments (MUNIZ and LEIVESTAD 1979, LEIVESTAD and MUNIZ 1976). These observations may be dependent on physiological stress phenomena, which means that surplus energy for growth no longer can be provided.

Humic substances, here measured as water colour, are known to have a strong complexing and detoxifying capacity towards metals and other ions. The distribution of the bigger trout in the brownest streams was interpreted as an effect of reduced toxicity of metals to trout and invertebrates (trout food).

IV. CONCLUSIONS

Correlations of data in this investigation showed that the trout abundance decreased at low pH (high H^+ ion concentration), low electro-conductivity and low alkalinity. The trout growth was favoured when abundance decreased, probably because of decreased competition for food and space. High water colour had a positive influence on trout growth, independent of the abundance, probably because of the metal-complexing and detoxifying ability of the humic substances.

This study showed that a pH exceeding 6.5 during summer favoured trout abundance. Referring to lowered pH values during spring and trout hatching, reproduction failure might be the reason for low abundance or complete disapperance of trout in the streams.

V. ACKNOWLEDGMENT

Many thanks to HÅKAN WICKSTRÖM at the Institute of Freshwater Research, Drottningholm, for assistance in the age determination of the trout.

VI. REFERENCES

ALMER, B., W. Dickson, C. EKSTRÖM and E. HÖRNSTRÖM. 1978. Sulfur pollution and the aquatic ecosystem. p. 271—311. *In* Sulfur in the environment. Part II. Ecological impacts. Ed.: J. O. Nriagu. John Wiley & Sons, New York.

BEAMISH, R. J., W. L. LOCKHART, J. C. VAN LOON and H. H. HARVEY. 1975. Long-term acidification of a lake and resulting effects on fishes. Ambio 4: 98—102.

BRUCE, J. P. and R. H. CLARK. 1969. Introduction to hydrometeorology. A. Wheaton's Co., Exeter. Great Britain. Pergamon Press Ltd.

DICKSON, W. 1979. Exempel på metalltoxicitet vid försurning och kalkning. Aquannalen (Soc. Aquat., Lund) 1: 2—7. (In Swedish.)

DRISCOLL JR, C. I., J. P. BAKER, J. J. BISOGNI JR and C. L. SCHOFIELD. 1980. Effects of aluminium speciation on fish in dilute acidified waters. Nature, Lond. 284: 161—164.

JENSEN, K. W. and S. E. SNEKVIK. 1972. Low pH levels wipe out salmon and trout populations in southernmost Norway. Ambio 1: 223—225.

LEIVESTAD, H., G. HENDREY, I. P. MUNIZ and E. SNEKVIK. 1976. Effects of acid precipitation on freshwater organisms. p. 87—111. *In* Impact of acid precipitation on forest and freshwater ecosystems in Norway. Ed.: F. R. Braeckke. SNSF-project. Res. Rep. 6/76 Oslo—Ås.

— and I. P. MUNIZ. 1976. Fish kill at low pH in a Norwegian river. Nature, Lond. 259: 391—392.

McDONALD, D. G., E. HOBE and C. M. WOOD. 1980. The influence of calcium on the physiological responses of the rainbow trout, *Salmo gairdneri*, to low environmental pH. J. exp. Biol. 88: 109—131.

McWILLIAMS, P. G. 1980 a. Effects of pH on sodium uptake in Norwegian brown trout (*Salmo trutta*) from an acid river. J. exp. Biol. 88: 259—267.

— 1980 b. Acclimation to an acid medium in the brown trout *Salmo trutta*. J. exp. Biol. 88: 269—280.

— and W. T. W. POTTS. 1978. The effects of pH and calcium concentrations on gill potentials in brown trout, *Salmo trutta*. J. comp. Physiol. 126: 277—286.

MUNIZ, I. P. and H. LEIVESTAD. 1979. Langtidsexponering av fisk til surt vann. Forsøk med bekkerøje *Salvelinus fontinalis* MITCHILL. SNSF-project IR 44/79 Oslo—Ås. (In Norwegian with English summary.)

ROSSELAND, B. O. 1980. Physiological responses to acid water in fish. 2. Effects of acid water on metabolism and gill ventilation in brown trout, *Salmo trutta* L., and brook trout, *Salvelinus fontinalis* MITCHILL. p. 348—349. *In* Ecological impact of acid precipitation. Proc. Inst. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

— I. SEVALDRUD, D. SVALASTOG and I. P. MUNIZ. 1980. Studies on freshwater fish population — effects of acidification on reproduction, population structure, growth and food selection. p. 336—337. *In* Ecological impact of acid precipitation. Proc. Inst. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

SWEDISH STANDARD METHODS: Nos 55028124, 55028123, SS 028122-2 and SS 028139. SIS, Box 3295, S-103 66 Stockholm, Sweden. (In Swedish.)

SYKORA, J. L., E. J. SMITH, M. SYNAK and M. A. SHAPIRO. 1975. Some observations on spawning of brook trout (*Salvelinus fontinalis* MITCHILL) in lime neutralized iron hydroxide suspensions. Wat. Res. 9: 451—458.

WRIGHT, R. F. and E. SNEKVIK. 1978. Acid precipitation: Chemistry and fish populations in 700 lakes in southernmost Norway. Verh. internat. Verein. Limnol. 20: 765—775.

Experiments with Brown Trout (*Salmo trutta* L.) during Spring in Mountain Streams at Low pH and Elevated Levels of Iron, Manganese and Aluminium

PAUL ANDERSSON [1] and PER NYBERG [2]

[1] The National Swedish Environment Protection Board, S-171 25 Solna, Sweden.

[2] Institute of Freshwater Research, S-170 11 Drottningholm, Sweden.

ABSTRACT

In situ experiments with brown trout were performed during acid spring episodes in 4 high-altitude mountain streams in central Sweden during 1982 and in one of the streams in 1983. The pH was measured daily from April 28—June 13 1982 and samples for other physical-chemical analysis were taken 5—9 times during this period. During the spring thaw the pH decreased from 6.1—7.2 to a minimum value of 4.5 in three of the streams. The mortality of the trout in these streams was high, particularly in the beginning of the period, when the pH was still above 5.5. The concentrations of Fe, Mn and Al were in the range of 550—1 200 (Fe), 80—180 (Mn) and 90—160 µg/l (Al) during periods of trout mortality. Gills from trout, both killed and surviving individuals, were analysed for these metals. Concentrations in the gills were 170—600 (Fe), 8—57 (Mn) and 2.5—41 (Al) mg/kg dw. Calculation of the hourly accumulation of metals in the gills showed that Fe was accumulated about 10 times faster than Mn and Al, which had about equal rates of accumulation. Concentrations and accumulation rates were highest for all three metals during periods of high mortality. In 1983 exposed fish survived the beginning of the thaw in spite of maximum concentrations of 52, 3.9 and 0.45 mg/l of Fe, Mn and Al respectively in the water. These metals were apparently present as non-toxic compounds. Although the three metals may have an additive effect, as the gills are the target organ for all of them, iron was probably the primary cause of trout mortality.

I. INTRODUCTION

The acidification of fresh waters in North America and Scandinavia has resulted in increased concentrations of many metals and increased levels of dissolved fractions of metals in the water (DICKSON 1975, 1980, DRISCOLL *et al.* 1980, BAKER and SCHOFIELD 1982, BORG and ANDERSSON 1984). Many of these metals, like Al, Mn and Fe may be toxic to fish in the concentration range that often occurs in acidified waters. There is now a recent comprehensive literature on the toxicity of aluminium (BAKER and SCHOFIELD 1980, 1982, DRISCOLL *et al.* 1980, MUNIZ and LEIVESTAD 1980, ROSSELAND and SKOGHEIM 1982, 1984, SKOGHEIM *et al.* 1984, SKOGHEIM and ROSSELAND 1984). Much work has also been devoted to studying the toxicity of iron (AMELUNG 1982, BRENNER *et al.* 1976, LUKOWICZ 1976, RAY and DAVID 1962, SYKORA *et al.* 1972, SMITH and SYKORA 1976). There are however, relatively few studies on the toxicity of manganese and its toxicity to fish is still a matter of some controversy (AGRAWAL and SRIVASTAVA 1980, NIX and INGOLS 1981).

The acute toxic action of all three metals seems at least partly to be related to metal precipitating on the gills, which leads to death by suffocation (LUKOWICZ 1976, MUNIZ and LEIVESTAD 1980, NIX and INGOLS 1981). As acidified waters often contain elevated levels of all these elements at the same time (BJÄRNBORG 1983, DICKSON 1980, DRISCOLL *et al.* 1980) the effect of the metals is additive.

The aim of this investigation was to follow the spring flood in a remote area with a fairly high acid loading, where a large fraction of the precipitation had accumulated as snow. The analysis of stream water and experiments with brown trout (*Salmo trutta*) in cages were performed to try and explain the dramatic decreases in the fish populations that have been observed in streams in the area during recent years.

Fig. 1. Location of the studied streams.

II. THE STUDY AREA

The four study streams are situated in central Sweden 530—800 m above sea level. The bedrock consists of granites in the lower parts, and sparagmites and quartzites at higher altitudes. Bogs and swamps dominate the upper parts of the catchment areas. The sizes of the drainage areas of the four streams Bjursvasslan brook, Djursvasslan brook, the Djursvasslan river and the Glötån river (Fig. 1), are in the range of 20—100 km^2. The first three are forest and mountain streams with no lakes in their catchment areas, while the Glötån river is the largest and has a couple of lakes in its drainage area.

All the streams, except the Glötån river, drain into the impounded Lake Lofssjön (Fig. 1). Various types of fishery management measures were discussed in the mid 1970s for the lake and its catchment area. Water quality and the composition of the fish fauna were therefore studied in a number of lakes and streams in the catchment. Bjursvasslan brook, Djursvasslan brook and the Djursvasslan river were in 1977 inhabited by brown trout (*Salmo trutta*), roach (*Rutilus rutilus*), minnow (*Phoxinus phoxinus*) and burbot (*Lota lota*) (Olofsson pers.comm.). During the spring flood of 1978, a great number of these fishes migrated downstream, and were caught in a stationary trap in the Djursvasslan river where they died soon after capture (Olofsson pers. comm.). In 1975—77 the pH of the snow was close to 5 and the pH of the Djursvasslan river did not change much during the spring thaw (Fig. 2). In 1978, however, the pH of the snow was very low and the thaw caused a severe drop in the pH of the stream. The fish were evidently trying to escape the toxic water quality. Since then, the pH of the snow has been in the range of 4.2—4.7 and the spring thaws have caused acid episodes (Fig. 2).

The abundance of brown trout in Bjursvasslan brook, Djursvasslan brook and the Djursvasslan river in 1977 was 6, 14 and 10 individuals per 100 m^2 respectively (Olofsson pers.comm.). Corresponding values from the same sites in 1981 were 0.3, 14 and 1 individuals (Gydemo pers. comm.). Moreover, roach, which had been numerous in 1977, were totally absent in 1981 (*op.cit.*) and reproducing populations of trout only existed in Djursvasslan brook, which had the highest pH (Fig. 2).

III. MATERIAL AND METHODS

Two parallel cages, each containing 10 hatchery-reared yearlings of brown trout (*Salmo trutta*), were placed in each of the four streams in 1982 (Fig. 1). The experiments started on April 28 just before the spring flood and were run continuously until June 13. The trout were exposed during 10 days or until all fish were dead. After 10 days of exposure or as soon as 50 per cent of the trout were dead, a new experiment was started. In the 1982 experiments, the trout were delivered from a local hatchery about 50 km south of the study area on three occasions and were kept in a large cage in the Glötån river before future experiments. In the experiment in Bjursvasslan brook in 1983, wild trout 10—12 cm in size from the adjacent Lofsån river (Fig. 1) were used. 10 fish in each cage were exposed from April 14 onwards, until their death. In the 1983 experiments one of the cages was kept in a plastic bag and the in-

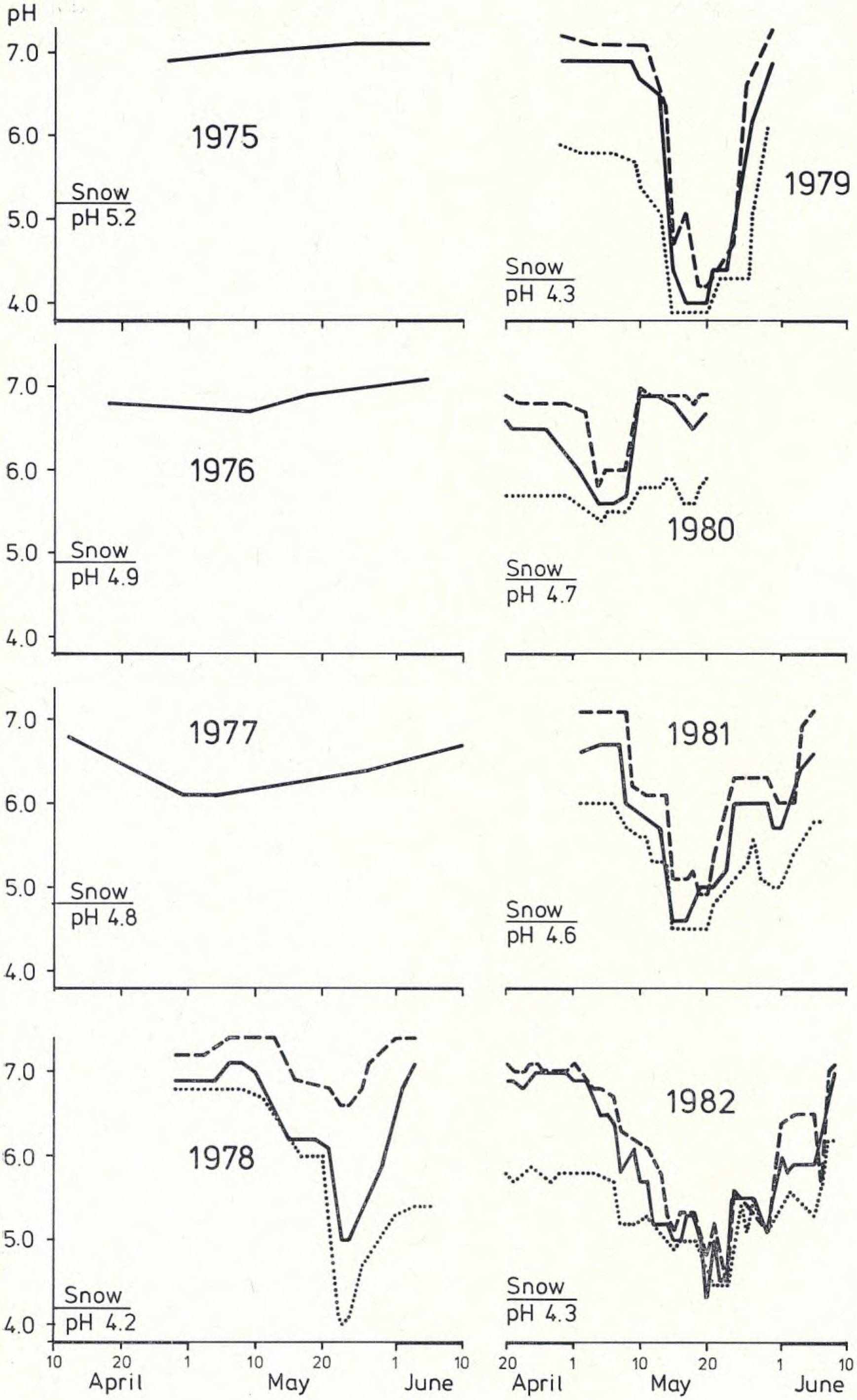


Fig. 2. The pH of snow 1975—82, in Bjursvasslan and Djursvasslan brooks 1978—82 and the Djursvasslan river 1975—82.

— — — — Djursvasslan brook, ——— Djursvasslan river, · · · · Bjursvasslan brook flowing water was filtered through several layers of commercial gauze bandage. The cages were inspected twice a day in 1982 and usually daily in 1983.

In each experiment in 1982 at least one sample of 3—5 trout with the same exposure time were taken for metal analysis of whole gills. On two occasions 15 trout from the Bjursvasslan brook and the Glötån river were separated into 3 subsamples to test the variation in metal concentration in the gills between groups of fish with the same exposure time.

Dead or killed trout were kept frozen at $-20°C$ prior to dissection. The whole gills were dissected, freeze dried and analysed for Fe, Mn and Al by graphite furnace atomic absorption spectroscopy (AAS). Sample preparation, digestion and determination of the metals were performed according to Borg *et al.* (1981). When calculating the rate of accumulation, the average concentrations of metals found in the three samples of trout gills from the Glötån river were used as background levels. The accumulation rates were calculated as the difference between the concentration in gills in each experiment and the background level, divided by the exposure time in hours.

Water samples for pH measurements were taken twice a month from 1975—77 in the Djursvasslan river. All figures in this study from 1978 and onwards are based on daily pH measurements during the periods studied. Water samples for analysis of water colour, Ca, Fe, Mn and Al were taken in Bjursvasslan brook, Djursvasslan brook, the Djursvasslan river and the Glötån river on 8, 5, 8 and 9 occasions respectively from April 20 — June 18, 1982. In 1983, the corresponding samples were taken on 10 occasions in Bjursvasslan brook from February 27 — June 14.

Samples for analysis of pH, colour, Ca and Al were taken in polyethylene bottles. Measurements of pH, and in 1983 also of water colour, were performed on the day of sampling and the other analyses were usually performed within two weeks. The Fe- and Mn-samples were taken in acid-cleaned polypropylene bottles, preserved with ultrapure nitric acid (2 ml/l), frozen at $-20°C$ and analysed within a year. The speciation of Al was investigated on four occasions in 1982 and on three occasions in 1983 by using an ion exchange technique (Driscoll 1980).

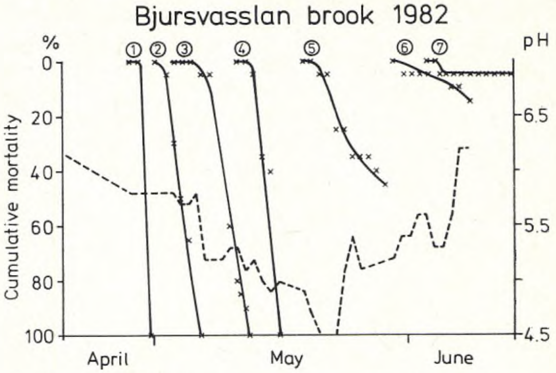


Fig. 3. The pH and cumulative mortality of brown trout in Bjursvasslan brook, April—June 1982.
x———x Cumulative mortality, — — — — pH,
① Experiment number

The pH was measured colourimetrically from 1975—81 and potentiometrically from 1981 and onwards. Water colour was measured colourimetrically during the whole period. Concentrations of Al were determined spectrophotometrically while Ca, Fe and Mn were determined by flame atomic absorption spectroscopy (AAS) according to Swedish Standard Methods (1981).

IV. RESULTS

Mortality and pH

In the first experiment in 1982 in Bjursvasslan brook, all of the brown trout were dead within 2 days (Fig. 3) and 50 per cent mortality was reached within 40 hours (Table 1). The mortality was high during the following three experiments (May 1—14) when 50 per cent of the trout died within 71—113 hours (Fig. 3, Table 1). However, during the last three experiments (May 19 — June 13), the survival of the trout increased and the cumulative mortality during 10 days of exposure was 45, 15 and 5 per cent respectively (Fig. 3).

In mid April, before the thaw, the pH value was around 6.2. It decreased later on, but remained fairly stable and in the range of 5.7—5.8 during experiments 1 and 2 (Fig. 3). After May 6, the pH decreased step-wise down to 4.5

Table 1. *Date of the experiments and average time for 50 per cent mortality in the four streams studied in 1982.*

Stream	Experiment number	Date	Number of hours for 50 % mortality
Bjursvasslan brook	1	April 28—April 30	40
	2	May 1—May 6	71
	3	May 5 —May 10	113
	4	May 11—May 14	88
	5	May 19—May 29	> 240
	6	May 30—June 9	> 240
	7	June 3—June 13	> 240
Djursvasslan brook	1	April 28—May 8	> 240
	2	May 9—May 19	> 240
	3	May 19—May 29	> 240
	4	June 3—June 13	> 240
Djursvasslan river	1	April 28—May 1	144
	2	May 3—May 13	242
	3	May 8—May 18	232
	4	May 19—May 29	> 240
	5	June 3—June 13	> 240
Glötån river	1	April 28—May 8	> 240
	2	May 8—May 19	> 240

on May 21—23 and then rose again to 6.2 on June 6.

The survival of trout in Djursvasslan brook was much better, and mortality was only noted during experiments carried out from April 28 — May 8 (10 per cent) and May 9—19 (25 per cent) (Fig. 4). The pH before the thaw was high (7.3) but it decreased nevertheless to 4.5 in late May. It then increased again to 7.1 in the beginning of June (Fig. 4).

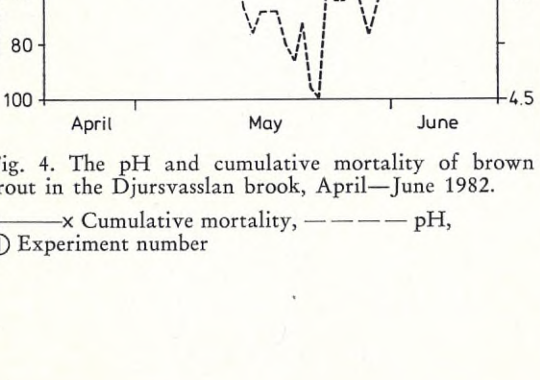


Fig. 4. The pH and cumulative mortality of brown trout in the Djursvasslan brook, April—June 1982.

x——x Cumulative mortality, — — — — pH,
① Experiment number

In the Djursvasslan river, which carries a mixture of water from Bjursvasslan and Djursvasslan brooks, mortality was highest during the first experiment, when 50 per cent of the trout were dead within 144 hours (Fig. 5, Table 1). The fish survived somewhat longer in the second and third experiments (May 3—18), but all of the fish died soon after the first deaths had occurred (Fig. 5). 50 per cent were dead in about 10 days (Table 1). None of the trout exposed in the stream from May 19 — June 13 died.

Even in this stream, the pH was high (7.0) at the time when the experiments started, but dropped during the following 22 days to 4.5 and then rose again to 7.0 at the end of the experimental period (Fig. 5).

The Glötån river is well buffered and has lakes in its catchment area. The lowest pH measured was 6.3. The mortality was negligible (5 per cent) during the two experiments (April 28 — May 19). Only one single trout was found dead (Fig. 6).

The brown trout from the Lofsån river, exposed in the 1983 experiments in Bjursvasslan brook, survived longer than the hatchery reared trout in the 1982 experiments. In contrast to the previous year the fish survived the first phase of the thaw in both unfiltered and filtered water. Although

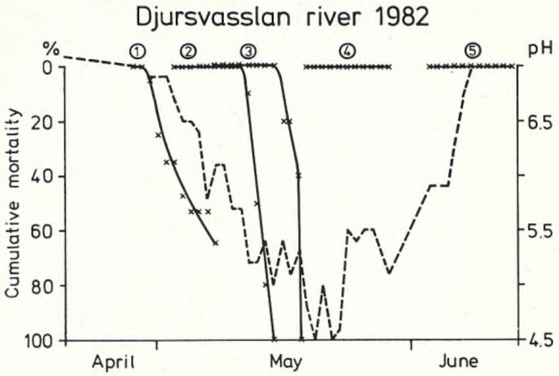


Fig. 5. The pH and cumulative mortality of brown trout in the Djursvasslan river, April—June 1982.

x——x Cumulative mortality, — — — — pH,
① Experiment number

the fish started to die at about the same time (May 9—12) in both cages, all fish died more rapidly in unfiltered water (Fig. 7). In the cage with unfiltered water, 50 per cent mortality was reached within ca 29 days compared to ca 39 days in the cage with filtered water. During the 1983 experiment the pH dropped from 6.7 (April 15) to 4.7 (May 2), stabilized around 4.6—4.7 for the next 25 days and then rose again to 5.4 in early June.

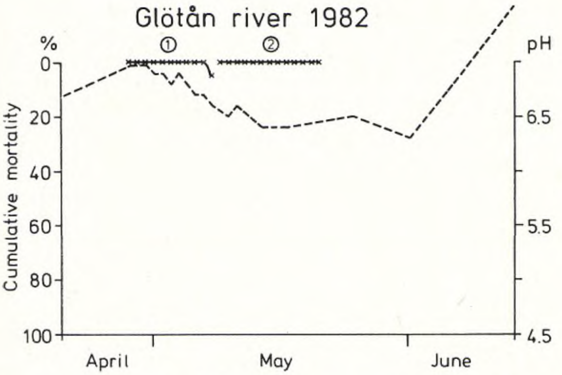


Fig. 6. The pH and cumulative mortality of brown trout in the Glötån river, April—June 1982.

x——x Cumulative mortality, — — — — pH,
① Experiment number

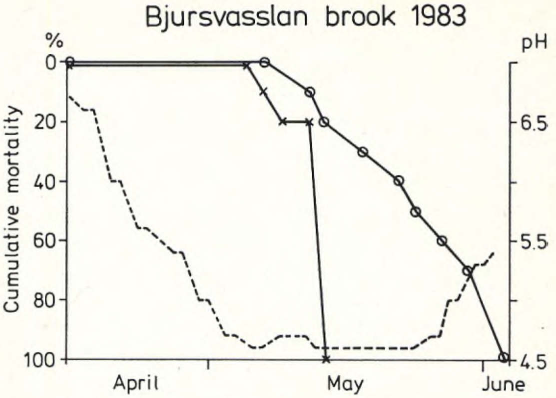


Fig. 7. The pH and cumulative mortality of trout in filtered and unfiltered water in Bjursvasslan brook, April—June 1983.

——x—— Cumulative mortality in unfiltered water, — — — — pH, ——⊖—— Cumulative mortality in filtered water

Water colour and concentrations of metals in water

In 1982, the concentration of total aluminium was about the same in all the streams and remained fairly stable during the period April 20 — June 1, although peak values of 140—150 µg/l were recorded in Bjursvasslan brook at this time (Fig. 8). The water colour in the streams was in the range of 90—150 mg Pt/l during the thaw (Fig. 8).

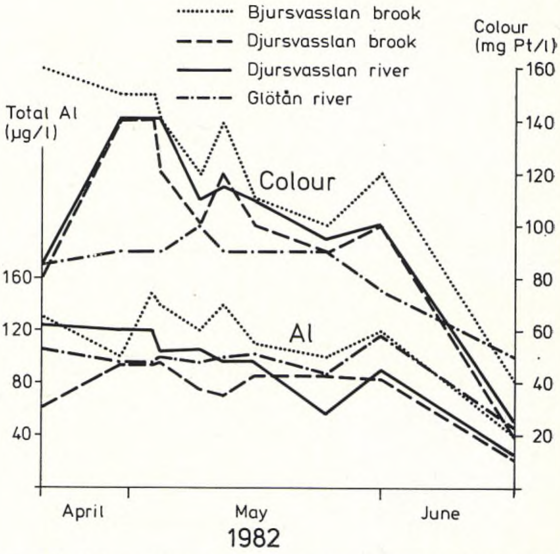


Fig. 8. Water colour and concentrations of aluminium in the four streams studied, April—June 1982.

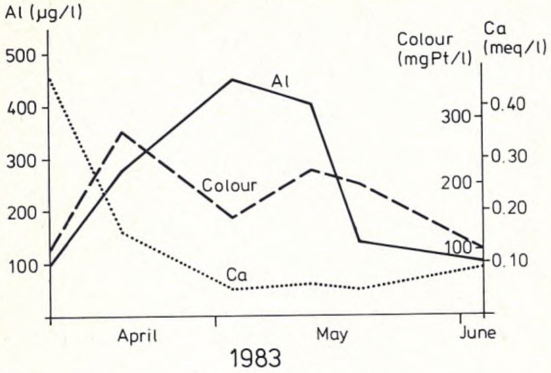


Fig. 9. Water colour and concentrations of calcium and aluminium in Bjursvasslan brook, April—June 1983.

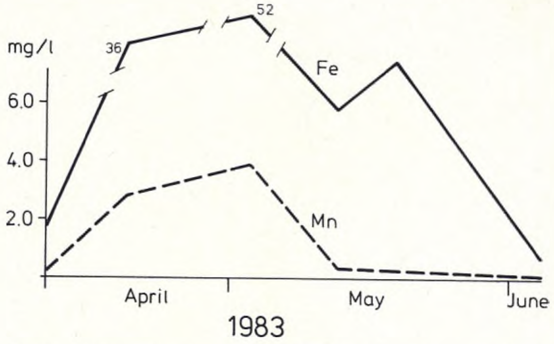


Fig. 11. Concentrations of iron and manganese in Bjursvasslan brook, April—June 1983.

Aluminium occurred mainly in suspended or organically bound forms. The fraction of labile inorganic aluminium was generally less than 10 % of total-Al at pH > 5.5, even during periods of high fish mortality. When the pH decreased below 5.5, this fraction increased to 25—30 % of total-Al, but the concentrations were still < 30 µg/l (Bjärnborg pers.comm.).

At the start of the study in Bjursvasslan brook in 1983, the aluminium concentration was about the same as the previous year, but it reached peak values of 398 and 449 µg/l in May (Fig. 9). The water colour increased from 100 to 280 mg Pt/l at the beginning of the thaw but decreased during the period of greatest run off from May 15—20 and ranged from 150—220 mg Pt/l (Fig. 9).

The fractions of Al showed a pattern similar to that found in 1982. The concentration of labile inorganic aluminium showed, however, a maximum value of 55 µg/l at pH 4.7. (Bjärnborg pers.comm.).

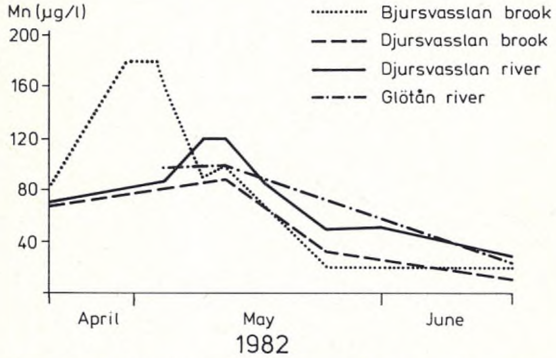


Fig. 10. Concentrations of manganese in the four streams studied, April—June 1982.

In 1982, the highest concentrations of manganese were measured in Bjursvasslan brook from the end of April to the beginning of May. The peak values of 180 µg/l then dropped rapidly and manganese concentrations were generally less than 100 µg/l in all the streams (Fig. 10).

The level of manganese in Bjursvasslan brook prior to the spring thaw in 1983 was comparable to the levels of 1982. In 1983, however, we observed a several-fold increase at the beginning of the spring runoff, when concentrations of 2.9 and 3.9 mg/l were measured in April and early May (Fig. 11).

The concentration of iron was high (1.2 mg/l) in Bjursvasslan brook in 1982 before and during the first phase of the thaw. In Djursvasslan brook and the Djurvasslan river, values were above 0.7 mg/l until mid May (Fig. 12). During the period of maximum discharge the Fe concentrations were 0.13—0.45 mg/l in all the four streams. The Fe concentration then rose again in the Djursvasslan river and Bjursvasslan brook, while it decreased in the other two streams (Fig. 12).

On April 10, 1983 the iron level in Bjursvasslan brook was similar to the previous year. However, it increased several-fold at the beginning of the spring runoff when concentrations of 36 and 52 mg Fe/l were measured in mid April and early May. These extremely high values then dropped, but the Fe concentrations were still as high as 5.8 and 7.4 mg/l in mid and late May (Fig. 11).

Before the thaw of 1982, the calcium concentrations were lowest in Bjursvasslan brook and the Glötån river (0.18 and 0.30 meq/l, respectively) and markedly higher in Djursvasslan brook and the Djursvasslan river (0.68 and 0.59 meq/l, respectively). During the thaw the calcium concentration decreased to 0.19 meq/l in the Glötån river. In the other three streams minimum values of 0.05—0.06 meq/l were noted (Fig. 13).

In 1983, the calcium concentration was higher (0.46 meq/l) in Bjursvasslan brook before the spring thaw, while the minimum values during the maximum runoff were in the same range as 1982 (0.05—0.06 meq/l) (Fig. 9).

Concentrations and accumulation rates of metals in gills

The concentration of iron in trout gills varied from 170—440, 220—290 and 230—600 mg/kg dw in Bjursvasslan brook, Djursvasslan brook and the Djursvasslan river, respectively, and in the Glötån river the iron concentration was 160—210 mg/kg dw ($\bar{x}=180$ mg/kg dw) (Table 2). The values were generally highest at the beginning of the experimental period. The concentrations of manganese and aluminium, which were much lower, were in the range of 8—57 and 2.5—41 mg/kg dw respectively. In general, there were no differences in the concentrations of any of

Table 2. *Date of experiments, hours of exposure and concentration of Fe, Mn and Al in gills of trout in 1982.*

Stream	Experiment number	Start of experiment (date)	Number of analysed trout	Hours of exposure in the stream	Me-concentration in gills (mg/kg dw)		
					Fe	Mn	Al
Bjursvasslan brook	1	April 28	5	57	280	35	15
	1	April 28	5	57	220	19	8.5
	1	April 28	5	57	330	21	14
					($\bar{x}=277$	25	13)
	2	May 1	3	96	440	17	9
	2	May 1	3	131*	360	25	8.5
	3	May 5	5	122*	240	26	8.5
	4	May 11	5	74*	180	19	10
	4	May 11	5	120*	210	16	6.5
	5	May 19	4	96*	170	8.6	2.5
	5	May 19	4	250	240	12	8.5
	6	May 30	5	248	210	16	6.0
	7	June 3	5	241*	180	11	8.0
Djursvasslan brook	2	May 9	5	227*	270	20	12
	2	May 9	5	243	280	20	12
	3	May 19	5	242	220	10	7.0
	4	June 3	5	242	290	11	22
Djursvasslan river	1	April 28	4	73*	600	57	24
	1	April 28	3	118	520	21	20
	1	April 28	5	236	400	23	24
	2	May 3	4	242*	380	41	36
	3	May 8	5	237*	460	45	41
	4	May 19	5	241	350	16	10
	5	June 3	5	240	230	8	6.0
Glötån river	1	April 28	5	240	170	12	7.0
	1	April 28	5	240	210	11	6.0
	1	April 28	5	240	160	12	5.0
					($\bar{x}=180$	12	6.0)

* Fish found dead in cage.

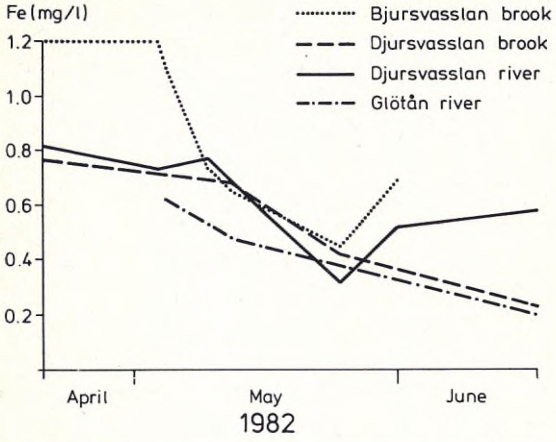


Fig. 12. Concentrations of iron in the four streams studied, April—June 1982.

these metals in samples from trout found dead in the cages and live trout from the same cages (Table 2).

The rate of accumulation of iron in the gills of trout exposed in Bjursvasslan brook was 1.35—2.30 mg Fe/kg dw. hour at the beginning of the period, but this later decreased to low values. The rates of accumulation of manganese, and aluminium in particular, were much lower and maximum values of 0.23 and 0.11 mg/kg dw. hour, respectively, were calculated (Fig. 14).

The highest rates of accumulation of all three metals were recorded in the gills of trout in the Djursvasslan river. During the first experiment, the calculated accumulations of Fe, Mn and Al were 5.75, 0.62 and 0.25 mg/kg dw. hour, respectively. The accumulation rates declined to their minima around May 7 but then increased again in the middle of the month (Fig. 14).

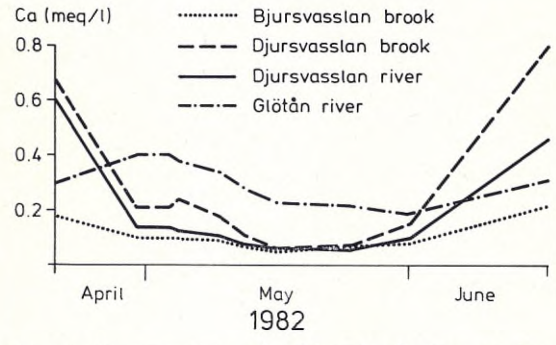


Fig. 13. Concentrations of calcium in the four streams studied, April—June 1982.

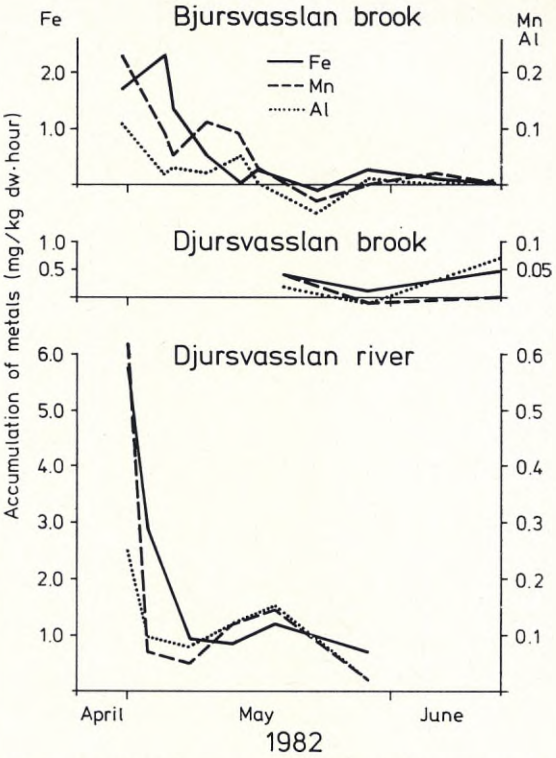


Fig. 14. Rates of accumulation of Fe, Mn and Al in gills of trout exposed in Bjursvasslan and Djursvasslan brooks and the Djursvasslan river in 1982.

Trout which had been caged in Djursvasslan brook had a low rate of metal accumulation in their gills during the entire study period (Fig. 14).

V. DISCUSSION

High mortality was observed among brown trout exposed in Bjursvasslan brook and the Djursvasslan river at the beginning of the spring thaw in 1982, while there was little or no mortality among trout exposed in Djursvasslan brook and the Glötån river. The heaviest mortality in the streams occurred when the pH was relatively high, but decreasing, at values above 5.5 (Figs. 3, 5). Relatively low mortality was registered in Bjursvasslan brook, Djursvasslan brook and the Djursvasslan river during the period of minimum pH (Figs. 3, 4, 5). In 1983, however, all brown trout survived during the first phase of the thaw in Bjursvasslan brook and mortality was not noted until May 7—12 at pH values of 4.6—4.7 (Fig. 7).

It has been shown that brown trout can survive at pH 4.6 in water with low levels of electrolytes and aluminium (LEIVESTAD *et al.* 1980). A high concentration of hydrogen ions is thus not a probable explanation for the high mortality in the beginning of the snow thaw of 1982.

Aluminium poisoning is often regarded as the main reason for the death of fish during acid episodes in natural waters (BAKER and SCHOFIELD 1980, 1982, DRISCOLL *et al.* 1980, MUNIZ and LEIVESTAD 1980, SKOGHEIM *et al.* 1984). It has been found that aluminium is most toxic at pH values around 5 (BAKER and SCHOFIELD 1980, FIVELSTAD and LEIVESTAD 1984, MUNIZ and LEIVESTAD 1980). We found, however, that the trout mortality was highest in Bjursvasslan brook and the Djursvasslan river in 1982 at pH values far above this level. In 1983, the mortality in Bjursvasslan brook was most severe at low pH values. Moreover only inorganic forms of aluminium have been shown to be toxic to fish (DRISCOLL *et al.* 1980, FIVELSTAD and LEIVESTAD 1984) and in our streams, the organic content, measured as water colour, was rather high and a large fraction of the total aluminium should have been bound in the form of organic Al-complexes, which are non toxic. This is illustrated by the Al/colour quotient, which was very low on all sampling occasions, except one, in Bjursvasslan brook during 1983 (Fig. 15), in spite of a fairly large variation in concentrations of Al (Fig. 9).

In 1982, the Al/colour quotient was 0.7—1.0 during the entire study period. During this year, however, water colour was not measured immediately after sampling, which makes these values uncertain. The few studies of the speciation of aluminium that were made in 1982, confirm, however, that most of the aluminium was bound in complexes (BJÄRNBORG pers.comm.). Even if the aluminium, which was generally measured as total-Al had been in inorganic forms, the concentrations of 1982 do not seem high enough to be acutely toxic to brown trout (MUNIZ and LEIVESTAD 1980). In 1983, however, a concentration of 55 µg/l of labile aluminium was measured (BJÄRNBORG pers. comm.) on the occasion with the highest Al/colour quotient. This sample was taken three days before the start of the fish deaths and aluminium may have contributed to the fish kills in 1983.

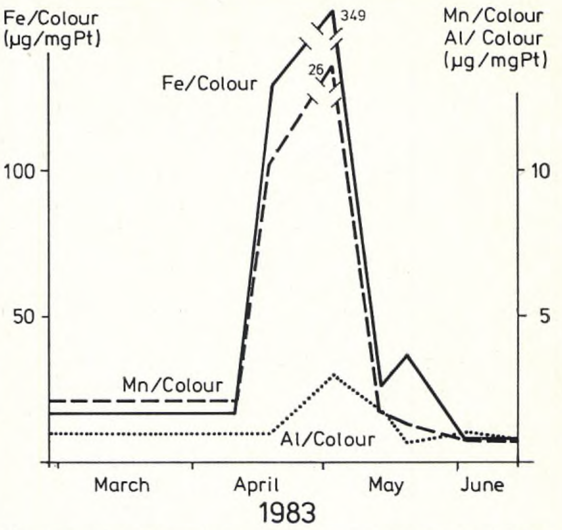


Fig. 15. Concentrations of iron, manganese and aluminium in relation to water colour in Bjursvasslan brook from February 27—June 14, 1983.

Little work has been done on the acute toxicity of manganese and the effects of different manganese compounds. AGRAWAL and SRIWASTAVA (1980) found a mortality of 30 per cent in the teleost *Colisa fasciatus* exposed to 2500 mg/l manganese as $MnSO_4$. NIX and INGOLS (1981) found a high mortality in rainbow trout (*Salmo gairdneri*) in a hatchery where the maximum concentration of Mn in the inlet water was 1.5 mg/l. They also found that the trout mortality was not directly correlated to the concentration of total-Mn, but rather to the concentration of oxidized manganese (*op.cit.*).

The concentrations of manganese recorded in the Djursvasslan river and Bjursvasslan brook in 1982 (ca 100—180 µg/l) thus do not seem acutely toxic to fish. In 1983, however, maximum values of 2.9 and 3.9 mg/l were recorded in Bjursvasslan brook (Fig. 11). Manganese is generally bound in organic complexes and may be non toxic, in spite of high total concentrations. In 1983 the Mn/colour quotient increased drastically from values around 2 before the thaw to ca 10 and 26 in mid April and early May (Fig. 15). These high quotients indicate that a certain fraction of the manganese occurred as inorganic ions or newly formed precipitates, which may be toxic to fish.

Iron in the form of Fe^{2+} has long been known to be harmful to fish and BRANDT (1938) regarded a concentration of 0.9 mg/l as toxic. LUKOWICZ (1976) found a very high mortality among rainbow trout at Fe^{2+} concentrations of 5 mg/l and AMELUNG (1982) noted 100 per cent mortality in larvae of the same species when they were exposed to 1.3 mg/l. SMITH and SYKORA (1976) were of the opinion that 0.97—1.27 mg/l may be the maximum concentration for successful hatching, survival and growth of coho salmon (*Oncorhynchus kisutch*) alevins.

In Bjursvasslan brook and the Djursvasslan river the concentrations of total-Fe were 0.65—1.2 and 0.56—0.77 mg/l respectively during the period of high trout mortality in 1982 (Table 1, Fig. 12). In 1983, the concentration of iron was very high (36—52 mg/l) in Bjursvasslan brook (Fig. 11). The brown trout survived these very high levels, but started to die when the concentrations decreased. The Fe/colour quotient was low (8—17) before and after the thaw, but increased to 129 and 349 during maximum discharge at the end of April (Fig. 15). This indicates that a certain amount of the iron may have occurred in inorganic and toxic forms. The trout started to die in the first half of May when the Fe/colour quotient was at a maximum.

The concentration of aluminium in the gills (2.5—41 mg/kg dw.) (Table 2) corresponds to 0.5—8.2 mg/kg ww., if one assumes the dry weight of gills to be 20 per cent of the wet weight. GRAHN (1980) found aluminium concentrations of 40—47 mg/kg ww in gills of dead cisco (*Coregonus albula*), SKOGHEIM *et al.* (1984) found concentrations of 70—341 mg/kg ww. in gills of dead or dying salmon (*Salmo salar*) and DICKSON (pers.comm.) found concentrations of 17.6 ± 3.5 mg/kg ww. in young sea-trout in a hatchery fed with aluminium-rich and limed inlet water. According to these results, the concentrations of aluminium in the gills exposed in this study are low.

The concentration of Mn in the gills was about the same as that of Al, while the concentration of Fe was in the range of 160—600 mg/kg dw. (Table 2) or 32—120 mg/kg ww. These concentrations, which partly reflect the fact that iron occurs naturally in blood and thus in gills, are more than ten times higher than those of Al and Mn. The concentrations of Fe in gills from trout exposed in the Glötån river were 160—210 mg/kg dw. or 32—42 mg/kg ww. If we assume these values to be representative for gills of healthy fish, as literature data are lacking, it implies that the increases in Fe concentrations were in the range of 0—80 mg/kg ww. The higher values in this range correspond quite well to the values cited above for aluminium levels in gills of dead or dying fish.

The rates of the accumulation of metals in gills were highest in Bjursvasslan brook and the Djursvasslan river in particular, during the first half of the experimental period when mortality was most severe (Fig. 14, Table 1). It is also obvious that the rate of accumulation of iron exceeded the accumulation rates of manganese and aluminium by an order of 10. If the assumed background concentrations of metals in gill samples from the Glötån river are incorrect, the absolute figures for the accumulation rates are biased. However, these values seem to be reasonable, because the accumulation of metals approached zero at the end of the study period, when water quality improved and mortality was negligible (Table 1, Fig. 14). The slightly negative accumulation values for the trout in Bjursvasslan brook may be due to reduced precipitation of metal hydroxides in gills in very acid water.

In spite of higher rates of metal accumulation, trout in the Djursvasslan river survived longer than in Bjursvasslan brook (Table 1). This may have been an effect of less pH stress, since the pH was generally higher in the Djursvasslan river during periods of high mortality (Figs. 3, 5). The slightly higher calcium concentrations in the Djursvasslan river compared to Bjursvasslan brook (Fig. 13) may also have favoured survival of the trout (BROWN 1983).

In the experiment which started around May 19, 1982 we recorded no mortality in the Djursvasslan river and Djursvasslan brook and relatively low mortality in Bjursvasslan brook (Figs. 3, 4, 5). This is noteworthy because there were only small changes in the concentrations of Fe as well as Mn and Al in the water during the experimental periods (Figs. 8, 10, 12). The pH was, however, very low (Figs. 3, 4, 5) and it has been shown that the toxicity of aluminium is reduced at low pH values (BAKER and SCHOFIELD 1982, FIVELSTAD and LEIVESTAD 1984). The calculated rates for the accumulation of metals were also low, or even negative, during periods when pH values were low (Fig. 14). Thus, increased survival at low pH values may have been caused by a change in ionic speciation and reduced metal sorption on the gills.

The results from the experiments in 1983 are different from the results of 1982. The brown trout survived during the first phase of the thaw (Fig. 7), in spite of very high concentrations of Al and in particular, Fe and Mn (Figs. 9, 11). The water colour was in the same range or slightly higher than the previous year (Figs. 8, 9). One explanation for the survival of these trout is that the metals were present as non-toxic compounds, *i.e.* bound to organic complexes (BAKER and SCHOFIELD 1980) or as precipitates of oxidized compounds (LUKOWICZ 1976). This is also indicated by the Me/colour quotients which were low prior to and at the beginning of the thaw (Fig. 15). Just before mortality was first noted the quotients for Mn/colour and Fe/colour had increased by an order of 10 (Fig. 15).

The prolonged survival of the brown trout after the rough filtration of the water (Fig. 7), indicates that the metal(s) was not only toxic when present as ions, but also in some particulate form, such as newly formed precipitates.

The effect of the three metals on the gills is probably additive, but based on the concentrations in the gills, we believe Fe to be the metal most responsible for trout mortality. However, this assumes that the three metals have about the same specific toxicity.

In 1982, the concentrations of Al and Mn were probably not high enough to cause acute mortality of brown trout. The concentrations of Fe, both in the water and in the trout gills, as well as the rate of accumulation of Fe in the gills, were about 10 times higher than those of Mn and Al. It is, however, conspicuous that the Mn/Fe quotients for the accumulation rates are more than twice as high for dead fish than for living trout. In 1983 the Me/colour quotients indicate that a certain fraction of all three metals occurred in inorganic and toxic forms. The increase in the Al/colour quotient was small and of short duration. On the other hand, the Fe/colour quotient reached maxima about 15 times higher than normal values and high values were recorded for a long period (Fig. 15).

The reason for the different water quality between 1982 and 1983 may depend on different hydrological conditions. In 1982, Fe and Mn were probably leached directly from the catchment area in reduced form. In 1983, however, the very high total concentrations of Fe and Mn and the high concentrations in relation to water colour during the thaw were probably caused by a flushing of the metals from mosses, algae and stones in the brook during high discharge. Thick layers of metal precipitates have also been observed in the brook during periods of low run off.

The fish kills during 1982 at relatively high pH-values were probably caused by newly leached, reduced forms of Fe and possibly Mn, while the kills during 1983, at low pH, were probably caused by redissolved forms of Fe and possibly Al from old precipitates in the stream.

The differences in trout mortality between the two years may have been affected by the long exposure time in 1983. It has also been shown that trout may be acclimatized to high levels of aluminium (FIVELSTAD and LEIVESTAD 1984, MUNIZ and LEIVESTAD 1980), which may explain why the wild trout were more tolerant to high levels of iron than the hatchery-reared trout in the 1982 experiments.

VI. CONCLUSIONS

Aluminium has been regarded as a primary toxic substance at low pH levels and a key factor behind fish kills in acidified waters. We cannot exclude the effects of Al in 1983, but our study indicates that iron and possibly Mn were the key metals involved.

One cannot exclude the possibility that trout mortality was due to some substance that was not analysed, such as organic compounds and other metals. Cd, Pb, Cu and Zn, were, however, only present in low maximum concentrations (BORG pers.comm.).

As the acidification of soil and ground water can be expected to result in greater leaching of iron and manganese, especially in environments low in oxygen, such as bogs, more attention should be paid to these metals in the future. It is also clear that an analysis of the total concentrations of these metals as well as an analysis of total aluminium tells us very little about their toxicity and the cause of fish mortality in acidified waters.

VII. ACKNOWLEDGMENTS

The authors are most indepted to Miss ELISABETH MELIN who prepared the gills. We would also like to thank Mr ERIK OLOFSSON and Mr GUNNAR OLOFSSON, who performed the field experiments and water samplings and also provided us with data of their own.

VIII. REFERENCES

AGRAWAL, S. J. and A. K. SRIWASTAVA. 1980. Haematological responses in a fresh water fish to experimental manganese poisoning. Toxicology 17: 97—100.

AMELUNG, M. 1982. Auswirkungen gelöster Eisenverbindungen auf die Ei- und Larvalentwicklung von *Salmo gairdneri* (RICHARDSON). Arch. Fisch. Wiss. 32: 77—87.

BAKER, J. P. and C. L. SCHOFIELD. 1980. Aluminium toxicity to fish as related to acid precipitation and Adirondack surface water quality. p. 292—293. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

— and C. L. SCHOFIELD. 1982. Aluminium toxicity to fish in acid waters. Wat., Air, Soil Poll. 18: 289—309.

BJÄRNBORG, B. 1983. Dilution and acidification effects during the spring flood of four Swedish mountain brooks. Hydrobiologia 101: 19—26.

BORG, H., A. EDIN, K. HOLM and E. SKÖLD. 1981. Determination of metals in fish livers by flameless atomic absorption spectroscopy. Wat. Res. 15: 1291—1295.

— and P. ANDERSSON. 1984. Fractionation of trace metals in acidified fresh waters by *in situ* dialysis. Verh. Internat. Verein. Limnol. 22: 725—729.

BRANDT, H. J. 1938. Die tötliche Menge an Gelöstem Eisen in Fischgewässern. Fisch. Ztg. 41: 593—595.

BRENNER, F. J., S. CORBETT and R. SHERTZER. 1976. Effect of ferric hydroxide suspension on blood chemistry in the common shiner, *Notropis cornutus*. Trans. Am. Fish. Soc. 105: 450—455.

BROWN, D. J. A. 1983. Effect of calcium and aluminium concentrations on the survival of brown trout (*Salmo trutta*) at low pH. Bull. Environ. Contam. Toxicol. 30: 582—587.

DICKSON, W. 1975. The acidification of Swedish lakes. Rep. Inst. Freshw. Res., Drottningholm 54: 8—20.

— 1980. Properties of acidified waters. p. 75—83. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

DRISCOLL, C. T. 1980. Chemical characterization of some dilute acidified lakes and streams in the Adirondack region of New York State. Doctor's Thesis, Cornell Univ.

— J. P. BAKER, J. J. BISOGNI JR and C. L. SCHOFIELD. 1980. Effect of aluminium speciation on fish in dilute acidified waters. Nature Lond. 248: 161—164.

FIVELSTAD, S. and H. LEIVESTAD. 1984. Aluminium toxicity to Atlantic salmon (*Salmo salar* L.) and brown trout (*Salmo trutta* L.): Mortality and physiological responses. Rep. Inst. Freshw. Res., Drottningholm 61: 69—77.

GRAHN, O. 1980. Fish kills in two moderately acid lakes due to high aluminium concentration. p. 310—311. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

LEIVESTAD, H., I. P. MUNIZ and B. O. ROSSELAND. 1980. Acid stress in trout from a dilute mountain stream. p. 318—319. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

LUKOWICZ, M. 1976. Der Eisengehalt im Wasser und seine Wirkung auf den Fisch. Fisch Umwelt 2: 85—92.

MUNIZ, I. P. and H. LEIVESTAD. 1980. Toxic effects of aluminium on the brown trout, *Salmo trutta* L. p. 320—321. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

NIX, J. and R. INGOLS. 1981. Oxidized manganese from hypolimnetic water as a possible cause of trout mortality in hatcheries. Progr. Fish Cult. 43: 32—36.

RAY, P. and A. DAVID. 1962. A case of fish mortality caused by precipitation of ferric iron in the River Daha at Siwan (North Bihar). Indian J. Fish. 9: 117—122.

ROSSELAND, B. O. and O. SKOGHEIM. 1982. Physiological stress and mortality of Atlantic salmon, *Salmo salar* L., in acid water with high levels of aluminium. Int. Council. Explor. Sea. C. M. 1982/M: 29. 15 p.

— and O. Skogheim. 1984. A comparative study on four salmonid fish species in acid aluminium-rich water. II. Physiological stress and mortality of one- and two-year-old fish. Rep. Inst. Freshw. Res., Drottningholm 61: 186—194.

Skogheim, O., B. O. Rosseland and I. Sevaldrud. 1984. Deaths of spawners of Atlantic salmon in River Ogna, SW Norway, caused by acidified aluminium-rich water. Rep. Inst. Freshw. Res., Drottningholm 61: 195—202.

— and B. O. Rosseland. 1984. A comparative study on four salmonid fish species in aluminium-rich water. I. Mortality of eggs and alevins. Rep. Inst. Freshw. Res., Drottningholm 61: 177—185.

Smith, E. J. and J. L. Sykora. 1976. Early developmental effects of lime-neutralized iron hydroxide suspensions on brook trout and coho salmon. Trans. Am. Fish. Soc. 105: 308—312.

Swedish Standard Methods (Svensk standard). 1981. SIS 0281. SIS, Box 3295, S-103 66 Stockholm. (In Swedish.)

Sykora, J. L., E. J. Smith and M. Synak. 1972. Effect of lime-neutralized iron hydroxide suspensions on juvenile brook trout (*Salvelinus fontinalis* Mitchill). Wat. Res. 6: 935—950.

Early Development of the Crayfish *Astacus astacus* L. in Acid Water

MAGNUS APPELBERG

Institute of Limnology, University of Uppsala, Box 557, S-751 22 Uppsala, Sweden

ABSTRACT

The sensitivity to acid environment within different stages of the reproductive cycle of *Astacus astacus* L. has been studied. When exposed to pH 5.0, a drastic loss of attached eggs were recorded within 30 days after oviposition. The mortality rate increased at the moment of hatching, and it was high during the early post-embryonal stages at low pH. The net accumulation of Na^+ and K^+ were depressed in embryos and hatchlings exposed to acid water, indicating disturbances of the ion-regulation mechanisms. Only small changes of the Ca^{2+} content was observed during the embryonal stage, while there was a rapid, pH-dependent, net accumulation of Ca^{2+} after hatching. No acclimation to acid environment was found after five weeks in the embryonal stage. It is proposed that reproductive failure may be one of the most important factors contributing to low abundance of *A. astacus* in acid waters.

I. INTRODUCTION

The effects of anthropogenic airborne acidification on natural waters is probably one of the most important factors, besides that of the fungal parasite *Aphanomyces astaci* SCHIKORA influencing the distribution of the crayfish *Astacus astacus* L. within their historical distribution area in Sweden (SVÄRDSON 1974, LEIVESTAD *et al.* 1976, FÜRST 1978, ALMER *et al.* 1978). Acid stress has been shown to cause haemolymph acidosis and failures in body salt regulation of crayfish (SHAW 1960, MALLEY 1980, DEJOURS and ARMAND 1980, MORGAN and MCMAHON 1982, MCMAHON and MORGAN 1983, APPELBERG 1984). Although adult intermoult crayfish may be tolerant to acute acid exposures (MORGAN and MCMAHON 1982, MCMAHON and MORGAN 1983), it is possible that chronic exposure to sublethal pH-levels adversely affects the crayfish. Neither APPELBERG (1979) nor MALLEY (1980) found, however, that pH above 5.0 had any primary effect on the mortality of *A. astacus* nor *Orconectes virilis* at moulting stage, although the uptake rate of Ca and the calcification of the exoskeleton was depressed.

In many fish species reproduction is strongly affected by low pH (JOHANSSON *et al.* 1977, TROJNAR 1977, PETERSON *et al.* 1980, BROWN and LYNAM 1981, RUNN and SOTHELL 1982 a, b), and as the reproduction cycle of Astacidae involves several stages that possibly could be sensitive to environmental changes, it may be affected similarly. This is also supported by the findings of FÜRST (1978) and FRANCE (1983), who have presented evidence indicating that early life history stages of crayfish may be more sensitive than others to acid stress. FRANCE (1983) recorded a negative effect of low pH on the number of attached eggs to O. *virilis*, and suggests that the cement used for egg-attachment and/or the external egg-membrane formation are deleteriously influenced by low pH. According to PANDIAN (1970), a rapid increase in ash weight, in eggs of lobsters, is followed by a simultaneous increase in water content, indicating a high net accumulation rate of salts towards the end of embryonal development. If the effects of acid stress on ion-regulation mechanisms during late embryonal development and early post-embryonal stages of *A. astacus* are comparable to those of adult crayfish, it is reasonable to believe that the effects may be more pronounced in the former, as has been demonstrated in early stages of fish development by RUNN and SOTHELL (1982 a, b).

In the present investigation some of the effects which acid stress may cause to both early embryonal and post-embryonal stages of the crayfish *A. astacus* have been studied and the results indicate that reproductive failure may be one of the most serious threats to the survival of *A. astacus* populations in acid lakes.

II. MATERIAL AND METHODS

Three different experimental trials were performed. In the first experiment, the effect of pH on the success of egg-attachment was studied, in the second, survival during embryonal development at different pH-levels, and in the third experiment, the effects of low pH during the last period of embryonal development and the first post-embryonal stages were studied with special reference to the net accumulation rates of Na^+, K^+, and Ca^{2+}. Throughout the study, the term "egg" will include both the embryo and the perivitelline fluid, as well as the egg-capsule.

Experimental animals

Mature *Astacus astacus* from the slightly acid Lake Rottnen, were used in the egg-laying experiment, berried females collected in the neutral, low-conductive River Ljungan, in the second experiment, and in the third experiment *A. astacus*, from Lake Tisnaren, were allowed to spawn in a local fish farm, where they were kept under natural conditions during the winter. The water of both Lake Tisnaren and the fish farm was neutral, with a relatively high Ca^{2+}content. Two weeks prior to the start of each experiment, the crayfish were placed in large plastic tanks, supplied with running tapwater at 10°C.

Experimental conditions

The aquarium system was composed of four units, with 132 l of water in each. An 80 l glass aquarium, placed in a water bath, was used as a reservoir for each unit. From each aquarium, water was recirculated to four 13 l plastic beakers, which were divided into two compartments by a fine-meshed net. Each compartment contained a single crayfish supplied with a shelter. The temperature was kept at 10 ± 1°C during experiment 1, while in the second experiment it was raised from 10°C to 17°C during the last 60 days, in order to simulate natural conditions, and in the third experiment it was kept constant at 15.0 ± 0.5°C.

Water quality

In experiments 1 and 2, distilled water mixed with tap water (10: 1) was used. Every fifth day, one tenth of the water volume was replaced with freshly mixed water, which had been allowed to equilibrate with the air, for three days after acidification, in order to reduce the amount of free CO_2. Conductivity varied between 12.5 and 17.5 mS m^{-1}, and concentrations of the major cations, measured during the experiment, were: Ca^{2+}0.45 mmol l^{-1}, Mg^{2+}0.10 mmol l^{-1}, Na^+0.17 mmol l^{-1} and K^+0.14 mmol l^{-1}. De-ionized water supplied with salts was used in the third experiment and the major ions, as added, were Ca^{2+}0.09 mmol l^{-1}, Mg^{2+}0.03 mmol l^{-1}, Na^+0.09 mmol l^{-1}, K^+0.015 mmol l^{-1}, SO_4^{2-}0.045 mmol l^{-1}, HCO_3^-, Cl^- and NO_3^- all 0.09 mmol l^{-1}, with the resulting conductivity measured as 4.8 mS m^{-1}. One half of the total water volume was exchanged every third day. All pH adjustments were made with addition of H_2SO_4 to the 80 l aquaria. In the first two experiments pH adjustments were made daily, while in the third experiment two automatic pH control devices, with combined gel-electrodes, were used, with pH kept within ±0.1 units.

Experimental performance

In the egg-laying experiment, 6 mature females were placed into each of three different media at pH 7.0, 6.0, and 5.0. Within 4 to 17 days, 15 of the females had extruded their eggs which were then counted on a regular basis for a period of 80 days. In experiment 2, groups of four egg-bearing females were placed in each of the four aquaria units one month after egg-laying. After an acclimation time of 14 days to the experimental water, pH was lowered to 4.5, 5.0 or 5.5 within 24 hours, and a control was kept at pH 7.8. The number and development of the eggs were then followed until hatching.

In the hatching experiment 24 females, with a minimum of 50 eyed eggs per female, were divided in three groups with 8 females in each. Each group was allowed to acclimate to the test water for 30 days, before the pH was lowered to 4.9 or 5.6 at a rate of 0.5 units h^{-1} in two of the groups. The control was kept at 7.0. On the first day after hatching half of the number of juvenile crayfish, from each pH-level, were placed into pH 4.9 or 7.0, in order to see if acclimation to an acid environment would be detected. Samples of eggs and juvenile crayfish were collected two times

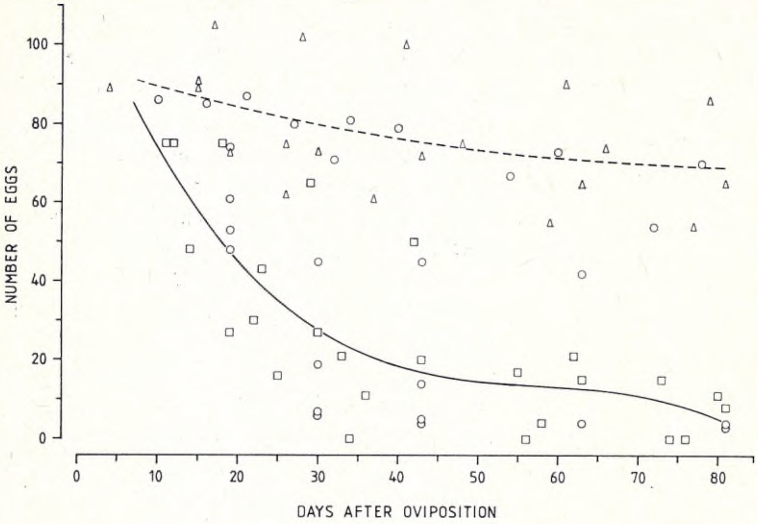


Fig. 1. Number of attached eggs to females of *Astacus astacus* during a period of 80 days after egg-laying. ○ pH 7.0 (n=6), △ pH 6.0 (n=4), □ pH 5.0 (n=5). Curves fitted by cubic regression at pH 6.0 and 5.0.

before hatching and four times within a three weeks post-hatching period. One week after which all juvenile crayfish had moulted once, the experiment was terminated.

Each sample, which contained a minimum of 6 individuals, was weighed wet after drying on filter paper for four seconds. Dry weights were measured and the water content calculated after freeze drying at −30°C for 24 hours. Weight values were obtained to nearest 1 µg using a CAHN 4700 electro balance. During post-embryonal stages, total lengths were measured (±0.1 mm) using a dissecting microscope fitted with an ocular micrometer.

Chemical analysis

Both eggs and juvenile crayfish were dissolved in 1 ml of 50 % HNO_3 for 24 h at 60°C. Measurements were made after diluting the samples in 20 ml of distilled water. All chemical analyses were performed using an atomic absorption spectrophotometer (UNICAM SP 1900). To both water and biological samples $CaCl_3$ was added prior to Ca^{2+} measurements to prevent phosphate binding and organic chelation. For the K^+ measurements, Na^+ was added to the standard solutions in order to reach an ion concentration comparable to the samples.

Statistical methods

Although eggs and hatchlings originating from different females are not strictly to be regarded as one statistical population, they were, however, treated as one population within all experiments. Unless specified, an unbalanced analysis of variance, two-way design with interaction, was used to test the significance of the effects of the independent variables (SAS Institute Inc., 1982). When needed, differences between least squares of means were used to separate different means. Only models with a high level of significance were used ($p < 0.001$, F-distribution).

III. RESULTS

Egg-attachment

A plot of the number of eggs laid at pH 5.0, 6.0, and 7.0 is shown in Fig. 1, and estimated curves, with regards to the decreasing number of eggs at pH 5.0 and 6.0, are fitted by cubic regression, from the time for oviposition until 80 days of embryonal development. Three out of six of the females in the control group (pH 7.0) lost most of their eggs within the first 30 days. Because the loss was regarded to depend on an experimental error, due to an intensive handling of the females, this group was not used in the statitstical analysis, nor is a regression line drawn, however, the values are shown.

When comparing the number of attached eggs at pH 5.0 and 6.0, during the 15 to 30 days after oviposition, it was found that there were significantly fewer attached eggs in the 5.0 group ($p < 0.01$, analysis of variance). During the suc-

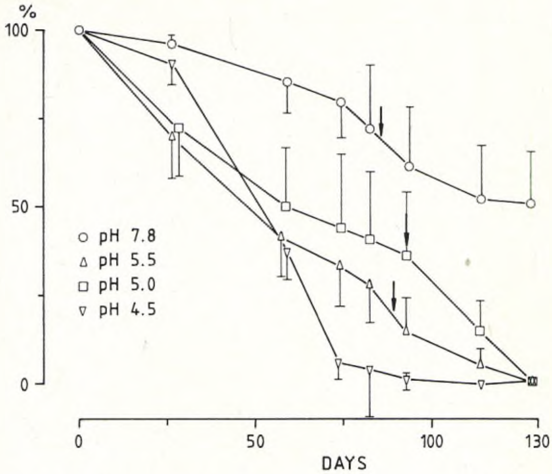


Fig. 2. Mean per cent survival by eggs of *Astacus astacus*, reared at four different pH-levels. Arrows indicate start of hatching. Mean ± S.E.M. n=4.

ceeding period, the pH 5.0 group lost most of their eggs, while the number of attached eggs in the pH 6.0 group showed only a small reduction. It was observed that the mucus, excreted by the cement glands, prior to egg extrusion, was more resilient and had a more yellow-whitish appearance in the pH 5.0 group than either pH groups 6.0 or 7.0.

Embryonal survival

There was a considerable difference between the survival of eggs reared at different pH-levels (Fig. 2). A two-factor analysis of variance with interaction, of the per cent decrease in number of eggs between each day of measurement, during prehatching period, shows that only differences between pH-levels are significant ($p < 0.05$). With respect to the separate pH-levels, the per cent decrease of eggs at pH 4.5, 5.0, and 5.5 were significantly different from that at pH 7.8 ($p < 0.05$). In all groups, heartbeats and developed embryos were observed, although the development seemed to be retarded at pH 4.5 compared to the other three groups during the same time period. In this group all eggs died before hatching, while hatching occurred in the other three groups. No hatchlings survived, however, the first moult at pH 5.0 nor at 5.5. Time of hatching was also

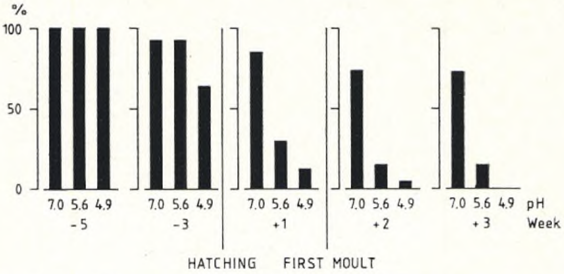


Fig. 3. Per cent survival by eggs of *Astacus astacus* exposed to three different pH-levels (7.0, 5.6 and 4.9) during a period of five weeks before hatching and three weeks past hatching.

found to be slightly delayed at low pH-levels ($p < 0.05$, Kruskal-Wallis non-parametric test).

Hatching and the first juvenile stages

In experiment 3, 8 egg-bearing females were placed in each of the three different media, at pH 4.9, 5.6, and 7.0, ca. five weeks before the predicted time for hatching. After hatching the juvenile crayfish were followed until they started to feed after their first moult.

Survival

A marked increase in mortality was recorded at the moment of hatching at both pH 4.9 and 5.6, with the largest portion of mortality, in both groups, due to hatching failure (Fig. 3). Apparently, as a result of the unsuccessful hatching, the female removed all partly hatched eggs within 24 hours. No juvenile crayfish, in the pH 4.9 group, survived longer than 5 days after their first moult.

Weight and length increase

The wet weight and percentage of water increased during both the pre- and posthatching periods until the time when the juvenile crayfish started to feed. A significant loss of dry weight was also noted during the same period ($p < 0.001$, Fig. 4 a and b). A slight depression of increase in wet weight was recorded at pH 4.9 and 5.6 compared to the pH 7.0 group during the prehatching period ($p < 0.05$), although no differences, with regards to pH, could be measured in neither dry weight nor percentage of water content.

Mean total lengths at both the first and second

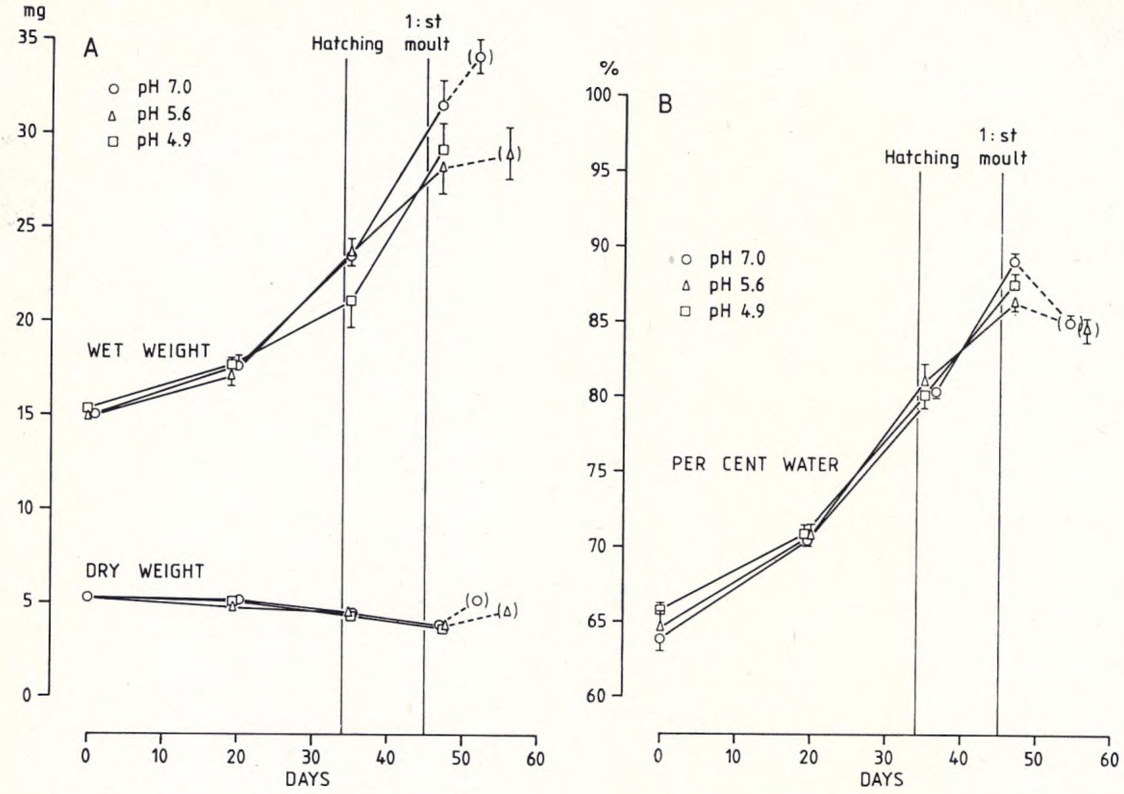


Fig. 4 A and B. Wet weight, dry weight and per cent water in *Astacus astacus* reared at pH 7.0, 5.6 and 4.9 during 34 days before hatching and 12 days past hatching. Mean ± S.E.M., n=8 during the prehatching period, n=6 during the posthatching period. Dots within brackets indicate that the juvenile crayfish have started to feed.

stage following hatching, are given in Table 1. Juvenile crayfish were ca. 5 % shorter at both stages at pH 4.9 compared to the other two groups ($p < 0.01$, analysis of variance).

Total content and net accumulation rates of ions

The total content and net accumulation rates of Na^+, K^+, and Ca^{2+} in eggs and juvenile crayfish in the third experiment, are illustrated in Fig. 5 a—c and Table 2, respectively. Effects of pH, time, and interaction between pH and time (*i.e.* the pH-effect on the net accumulation rates of ions) were tested by using an unbalanced two-factor analysis of variance with interaction for the pre- and posthatching period separately.

Prehatching

During the embryonal stage, there was a highly significant increase in both Na^+ and K^+ content

Table 1. *Total length of juvenile Astacus astacus, reared at pH 7.0, 5.6, and 4.9, in the first and second stage after hatching. Length in mm, S.E.M. within brackets, n=6.*

pH in media	7.0	5.6	4.9
first stage	8.37 (0.08)	8.44 (0.10)	8.01 (0.05)
second stage	11.17 (0.17)	11.03 (0.13)	10.53 (0.08)

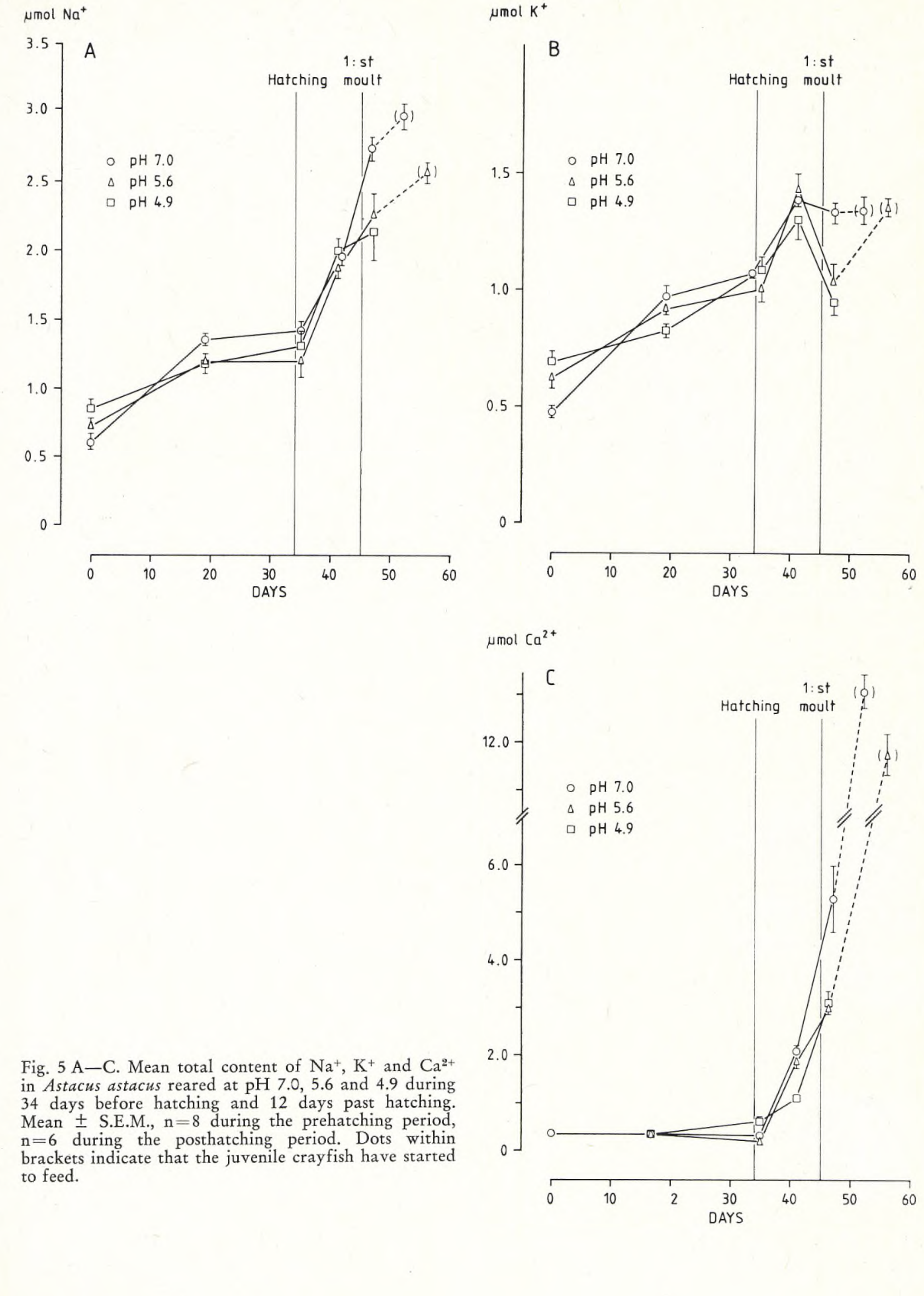


Fig. 5 A—C. Mean total content of Na^+, K^+ and Ca^{2+} in *Astacus astacus* reared at pH 7.0, 5.6 and 4.9 during 34 days before hatching and 12 days past hatching. Mean ± S.E.M., $n=8$ during the prehatching period, $n=6$ during the posthatching period. Dots within brackets indicate that the juvenile crayfish have started to feed.

Table 2. *Mean net accumulation rate of Na^+, K^+, and Ca^{2+} in Astacus astacus, obtained as mean changes between initial and last measurement in each of the 34 days of prehatching and the 12 days of posthatching periods.*

pH in media	Prehatching			Posthatching		
	7.0	5.6	4.9	7.0	5.6	4.9
nmol Na^+/h	1.0	0.5	0.5	8.2	3.6	2.9
nmol K^+/h	0.7	0.5	0.5	0.9	0.1	−0.5
nmol Ca^{2+}/h	0.0	−0.3	0.3	17.2	9.8	8.5

of the eggs ($p < 0.001$). The differences in net accumulation rates of these two ions were clearly pH-dependent ($p < 0.05$, and $p < 0.001$, respectively), with a depressed rate noted at low pH. No changes in Ca^{2+} content, during the first 19 days after the start of the experiment, were noted, although significant differences were found on the day following hatching (day 35). It is possible that the slight differences in time of sampling between the three groups, in combination with the drastic increase in net accumulation rate of Ca^{2+} after hatching, may constitute the main reason for these differences.

Posthatching

During the posthatching period, mean total contents of both Na^+ and K^+ were found to be pH-dependent ($p < 0.05$), while low pH caused a significant decrease in the net accumulation rate of K^+ only ($p < 0.01$). The net accumulation rate of Ca^{2+} increased rapidly after hatching and acid exposure was found to cause both a depression of mean total Ca^{2+} content and net accumulation rate ($p < 0.001$, respectively).

In order to compare the effect of acid stress during the prehatching period to posthatching ion contents, newly hatched juvenile crayfish, reared at pH 4.9 and 5.6 for 34 days as eggs, were placed in water of pH 7. Juvenile crayfish reared at pH 5.6 and 7.0 before hatching were also placed in pH 4.9. Samples were taken on the second day following the first moult (Table 3). In the statistical model, pH-levels during pre- and posthatching periods were used as independent variables.

Both the mean Na^+ and K^+ content were found to be dependent on prehatching pH ($p < 0.05$), but only the K^+ content was affected also by the posthatching pH ($p < 0.01$). The depression of the Na^+ content, due to low prehatching pH, increased by the interaction of low posthatching pH ($p < 0.01$).

The mean total content of Ca^{2+} on the second day after the first moult, on the other hand, was

Table 3. *Mean total content of Na^+, K^+, and Ca^{2+} in Astacus astacus on the second day after the first moult. After the embryonal period (34 days) at pH 7.0, 5.6, and 4.9, the newly hatched crayfish were transferred to pH 7.0 and 4.9 for 12 days. S.E.M. within brackets, $n=6$.*

pH in media during posthatching		pH in media during prehatching		
		7.0	5.6	4.9
umol Na^+	7.0	2.75 (0.09)	2.05 (0.10)	1.86 (0.11)
	4.9	2.01 (0.08)	1.95 (0.15)	2.14 (0.20)
umol K^+	7.0	1.33 (0.05)	1.04 (0.03)	1.01 (0.03)
	4.9	1.08 (0.07)	0.93 (0.09)	0.96 (0.07)
umol Ca^{2+}	7.0	5.30 (0.70)	4.06 (0.11)	3.43 (0.11)
	4.9	6.79 (0.39)	3.31 (0.29)	3.10 (0.23)

significantly influenced by the prehatching pH ($p < 0.001$), although only small changes in Ca^{2+} contents were recorded during the prehatching period (Fig. 5 c). It was also noted that similar to Na^+, a low pH during posthatching alone does not significantly reduce the mean Ca^{2+} content, although a low pH during both the pre- and posthatching period does.

IV. DISCUSSION

Experimental conditions

Differences in ion content, in ion ratios of the media, and also raised CO_2 levels due to acidification, are factors affecting results of experimental studies on freshwater organisms at low pH (NEVILLE 1979, BROWN 1981, MCWILLIAMS 1982, 1983). In the present study therefore, the different ion compositions of the three experimental designs may directly influence the results. As excess CO_2 was assumed to be eliminated by a three day acclimation of the test water in experiments 1 and 2, and the calculated maximal amount of free CO_2 in experiment 3 was low, ($pCO_2 < 2.5$ mm Hg), effects due to CO_2 may be regarded as negligible.

Oviposition

According to CHEUNG (1966), one vitelline membrane is present surrounding the ova at the time of oviposition in Astacidae. Following egg extrusion and the external fertilization in the brood pouch which has previously been filled with mucus secreted from the cement glands, this membrane combines with two other inner layers forming a "trichromatic", or fertilization membrane. The outermost membrane then forms the funiculus and attaches to the pleopod. According to CHEUNG (*op. cit.*), the cement for egg-attachment originates from the egg itself, as a consequence of fertilization, and AIKEN and WADDY (1980) have mentioned this as a possible explanation of why fertilized eggs of lobsters usually attach more successfully than unfertilized. A disruption of the fertilization process, due to an acid-induced reduction of the fertilization capacity of the externally deposited spermatophores, may be a possible explanation to the drastic loss of eggs during the first 30 days after oviposition at pH 5.0, recorded in this study. However, the rapid loss of eggs from three of the females at pH 7.0, indicates that the egg-attachment process is easily disturbed by factors other than low pH. The direct effect of low pH on the cement or formation of the funiculus may also be considered as important factors contributing to egg loss. It has earlier been suggested that the outer layer of the egg-membrane is epicuticular and secreted by the cement glands (see AIKEN and WADDY 1980, for review). FRANCE (1983) recorded a reduced elasticity of the funiculus and a decrease in the number of attached eggs to *Orconectes virilis*, in an experimentally acidified lake, and suggests that an incomplete hardening and calcification of the egg-membrane as the main reason for egg reduction.

Survival during hatching

Delays in time for hatching and partly hatched individuals, apparently not capable of successfully breaking through the egg-capsule at low pH, have been observed as results of acid stress to fish eggs (RUNN *et al.* 1977, PETERSON *et al.* 1980, BROWN and LYNAM 1981, NELSON 1982). It is suggested that a decrease in chorionase activity and changes in the physical structure of the outer mucopolysaccharide layer of the chorion are the main reasons for this delay and reduced hatching success (RUNN *et al.* 1977, HAYA and WAIWOOD 1981). DAVIS (1964) found that hatching of *Homarus* eggs were preceded by a rupture of the outer trichromatic membrane, in the cephalic region, indicating a weakening of the outer membrane which may be caused by enzymatic processes, which is also supported by PANDIAN (1970). PANDIAN (*op cit.*) also recorded a more than two-fold increase in egg-membrane dry weight of lobsters during the embryonal development, and it is possible that this membrane formation is affected by low pH. Several possible factors therefore, may be acting independently or collectively in contributing to the failure in hatching success at low pH recorded in this study: 1) a change in the structure of the egg-capsule, originating during the external membrane formation, 2) a reduced pressure within the embryo, due to a reduced ion uptake and ion concentration gradient, or 3) an obstruction of some unknown hatching enzyme. An increased metabolic rate, recorded during the moment of hatching in eggs of *A. astacus*, also suggests that the condition of the embryo may be of importance (Appelberg 1983).

Weight and length increase

Reduced growth rates as a result of low pH, have been reported to occur in both adult crayfish and fish (Leivestad *et al.* 1976, Rosseland 1980, Buck and Siewert 1980). The decrease in wet weight noted in the present study, is probably due to reduced salt and water uptake in the eggs. Nelson (1982), found a reduced length increment, in alevins of rainbow trout, to be a function of pH, and demonstrated a decrease of the amount of water-soluble protein in the yolk sac of acid-stressed alevins, suggesting a disturbance of protein metabolism, which also may be a plausible explanation to the slightly reduced length increment at pH 4.9 following the first moult found here.

Total ion content

In naturally developed *A. astacus*, Mackeviciene (1975) obtained an increase from 3.5 ‰ in the first stage to 16.9 ‰ of Ca^{2+} content (in relation to dry weight) after the first moult. The corresponding values at pH 7.0, from the third experiment in the present study, would be 1.9 ‰ and 10.2 ‰, respectively. The differences between the two studies may be attributed to differences in stages of moulting and also to the Ca^{2+} content of the water, as these two factors greatly affect calcium metabolism. Values for Na^+, K^+, and water content were also found to correspond with those of Mackeviciene (1975).

Changes in ion content

Increased accumulation rates of ions and water towards the end of the embryonal development has been recorded in eggs of lobsters (Pandian 1970) and the increase in water, Na^+, and K^+ content during the 34 days prior to hatching demonstrates that a similar accumulation occurs in *A. astacus*. This ion accumulation corresponds to the raised metabolic rate recorded during the last embryonal phase in eggs of *A. astacus* (Appelberg 1983). This increase, combined with the acid-induced depression of Na^+ and K^+ content, is comparable to the changes in Na and K found during the late embryonal development of brown trout recorded by Runn and Sothell (1982 b). During the prehatching period, the net accumulation rates of both Na^+ and K^+ were significantly lower at low pH, which suggests that acid stress acts on embryonal ion-regulation mechanisms in a similar manner as to that occurring in adult crayfish. (Shaw 1960, Morgan and McMahon 1982, McMahon and Morgan 1983, Appelberg 1984).

A decrease in intracellular K^+ concentration during acid stress, has been reported to occur in fish tissue (McDonald and Wood 1981, Fugelli and Vislie 1982) and also in haemocytes of *A. astacus* (Appelberg 1984). The suggestion that K^+ acts as an osmoeffector, preventing cell swelling during salt losses in the extracellular fluid, due to low pH, would explain the reduced K^+ content in both acid-exposed eggs and hatchlings. The decrease in K^+ content at pH 5.6 and 4.9, observed after the first moult, may be an effect of increased water uptake and low extracellular ion content originating during the ecdysis (Passano 1960).

There was a marked difference between the net accumulation rate of Ca^{2+} compared to that of Na^+ and K^+, during the embryonal stage, and apparently there was no embryonal calcification at any of the pH-levels. A low permeability to Ca^{2+} was found by Chaisemartin *et al.* (1969) for eggs of *Austropotamobius pallipes*, indicating that both influx and efflux rates were low. The drastic increase in Ca^{2+} net accumulation rate after hatching was depressed by low pH, which agrees with the findings of Malley (1980) who obtained an inhibition of the Ca^{2+} uptake rate at pH below 6.0 in postmoult *Orconectes virilis*. The principle mechanism behind this reduction is thought to be both a reduced exchange of internal K^+ with external Ca^{2+} and also a reduced concentration of HCO_3^- accompaning the Ca^{2+} uptake over the gills (Greenaway 1974, 1979, Malley 1980).

The Ca^{2+} uptake mechanism was obviously more affected by low pH during the 34 days of embryonal development than by low pH during the 12 days of post-embryonal development. These results suggest that acid exposure during late embryonal development may have disrupted the Ca^{2+} uptake mechanism, although only small changes in the total Ca^{2+} content of the eggs were recorded. If this hypothesized inhibition affects the efficiency of postmoult net accumulation of Ca^{2+}, it must also to some extent be irreversible, or at least recover at a slow rate. MALLEY (1980), on the other hand, found that the acid-induced inhibition of the Ca^{2+} uptake rate in adult *O. virilis* was partially a reversible process. It is not possible, however, to explain this contradiction with the results obtained in the present study.

MCWILLIAMS (1980) and RUNN and SOTHELL (1982 b), suggest that acid acclimated fish have a sodium transport capacity working below the saturation level, which increases when exposed to neutral pH. MCWILLIAMS (1983) also found that there was a less pronounced loss of bound Ca^{2+} from the gills of acid-acclimated brown trout at low pH, resulting in a smaller increase of the Ca^{2+}-dependent permeability to certain ions. Although no tendency towards acclimation was observed in the present study, it cannot be concluded that an adaption to acid environment does not occur in crayfish, since the origin of the crayfish, time spent in different media, and also the ionic content of the media may be of great importance.

Ecological implications

In Sweden, natural populations of *A. astacus* spawn in the middle of October and hatch towards the end of June or in July. During this period of embryonal development, there are often two pH minima. In slightly acidified lakes, one occurs in the autumn, as a consequence of heavy rainfall and run-off, and the other in spring, during snowmelt (ALMER *et al.* 1978). The high egg-loss rate during the time period following oviposition obtained in this study at pH 5.0, indicates that if low pH in autumn coincides with the oviposition of *A. astacus*, it may be one of the most serious threats to reproductive success. FRANCE (1983) recorded an increase in the number of partially berried females of *O. virilis* as pH decreased in an experimentally acidified lake.

A decrease in pH during spring, may have several detrimental consequences. In the present study, low pH during the last phase of embryonal development, resulted in a high mortality rate during hatching and a depressed net accumulation rate of ions, especially Ca^{2+}, during the early post-embryonal development. A low Ca^{2+} content in the exoskeleton may influence vulnerability to predation (STEIN 1975), but would not be a direct cause to death (APPELBERG 1979).

SVÄRDSON (1974) showed that the abundance of *A. astacus* was low in waters with pH less than 6.0, and findings from the present study indicate that failure in reproductive success may be one of the most important factors contributing to this low abundance.

V. SUMMARY

The effect of acid stress on reproductive stage of the crayfish *Astacus astacus* has been studied and it is shown that several stages of the reproductive cycle are sensitive to low pH.

(1) During egg-attachment there was a drastic loss of eggs at pH 5.0 within the first 30 days after oviposition. A disturbed fertilization process or an inhibitory effect by low pH on the outer egg-membrane formation and/or the cement used for egg-attachment are suggested as possible factors contributing to this loss.

(2) The mortality rate during the embryonal development was higher at pH 5.5 and below, compared to neutral water. At the moment of hatching, mortality increased drastically, and may be due to several factors interacting independently or collectively, a) acid-induced change in the structure of the egg-capsule, b) reduced pressure within the embryo or c) inhibition of some unknown hatching enzyme. A high rate of mortality was also recorded at low pH during the early post-embryonal stages.

(3) The slightly depressed rate of increase in wet weight at low pH is thought to be a result of disturbed ion regulation during the embryonal stage.

(4) Total ion content and net accumulation rates of Na^+ and K^+ within the embryos were affected by low pH. Acid stress during post-embryonal stages showed a significant effect on the total content of both Na^+ and K^+, but with respect to the net accumulation rate only K^+ was affected.

During embryonal development, only small changes of the Ca^{2+} content in the eggs were observed, while there was a drastic, pH-dependent increase in net accumulation rate after hatching.

(5) No acclimation to acid environment, during 34 days of embryonal development, was recorded after hatching. Since the Ca^{2+} contents, in hatchlings after their first moult, were more influenced by pH during prehatching than posthatching, it is suggested that there was, an at least partially irreversible, inhibition of the ion-regulatory mechanism of Ca^{2+}.

(6) The combined results from the three experiments of the present study, indicate that both egg-attachment and the hatching process, in combination with disturbances of the net accumulation rates of Na^+, K^+, and Ca^{2+}, may be highly sensitive to acid stress. It is proposed that reproductive failure may be one of the most important factors to the low abundance of *A. astacus* in acid waters in Sweden.

VI. ACKNOWLEDGMENTS

The author is greatly indebted to Professor B. Pejler, Dr P. Runn, Fk T. Odelström and MA Richard Johnson for their criticism of the manuscript, and to Isabella Jansson for technical assistance. Also Mr B. Andersson is thanked for preparing the illustrations. The study was financially supported by the National Swedish Board of Fisheries.

VII. REFERENCES

Aiken, D. E. and S. L. Waddy. 1980. Reproduction. p. 47—56. *In*: The biology and management of lobsters. Vol 1: Physiology and behavior. Eds: J. Stanley and B. E. Phillips. Academic Press, London.

Almer, B., W. Dickson, C. Ekström and E. Hörnström. 1978. Sulfur pollution and the aquatic environment. p. 271—311. *In* Sulfur in the environment: part II. Ecological impacts. Ed: J. O. Nriago. Wiley & Sons, New York.

Appelberg, M. 1979. The effect of low pH on *Astacus astacus* L. during moult. Paper presented in the second Scandinavian symposium on freshwater crayfish, Lammi, Finland, 1979. 12 p. (Mimeographed.)

— 1983. Response of acid stress upon the oxygen uptake in eggs of the crayfish *Astacus astacus* L. p. 59—70. *In*: Freshwater Crayfish V. Pap. Fifth Int. Symp. Freshw. Crayfish, Davis, Cal., USA. 1981. Ed.: C. R. Goldman. AVI Publishing Company, Westport, Conn.

— 1984. Changes in haemolymph ion concentration of *Astacus astacus* L. and *Pacifastacus leniusculus* (Dana) after exposure to low pH and aluminium. Hydrobiologia. (Submitted.)

Brown, D. J. A. 1981. The effects of various cations on the survival of brown trout *Salmo trutta*, at low pHs. J. Fish. Biol. 18: 31—40.

— and S. Lynam. 1981. The effect of sodium and calcium concentrations on the hatching of eggs and the survival of the yolk sac fry of brown trout *Salmo trutta* L. at low pH. J. Fish Biol. 19: 205—211.

Buck, J. and H. F. Siewert. 1980. Effects of low pH levels on body weight of crayfish. Proc. Indiana Acad. Sci. 89: 232—233. (Abstract.)

Chaisemartin, C., P. N. Martin and B. Echlancer. 1969. Equilibre thermodynamique calcique dans l'ovarie et dans l'oeuf chêz *Austropotamobius pallipes* Le. Cr. Seanc. Soc. Biol. 163: 775—776.

Cheung, T. S. 1966. The development of egg-membranes and egg attachment in the shore crab, *Carcinus maenas*, and some related decapods. J. mar. biol Ass. U.K. 46: 373—400.

Davis, C. C. 1964. A study of the hatching process in aquatic invertebrates. 8. Events of eclosion in American lobsters *Homarus americanus* Milne-Edwards. Am. Midl. Nat. 72: 203—210.

Dejours, P. and J. Armand. 1980. Haemolymph acid-base balance of the crayfish *Astacus leptodactylus* as a function of the oxygenation and the acid-base balance of the ambient water. Respir. Physiol. 41: 1—11.

France, R. L. 1983. Response of the crayfish *Orconectes virilis* to experimental acidification of a lake with special reference to the importance of calcium. p. 98—111. *In* Freshwater Crayfish V. Pap. Fifth Int. Symp. Freshw. Crayfish, Davis, Calif., USA 1981. Ed.: C. R. Goldman. AVI Publishing Company, Westport, Conn.

Fugelli, K. and T. Vislie. 1982. Physiological response to acid water in brown trout (*Salmo trutta*) L. cellvolume regulation in heart ventricle tissue. J. exp. Biol. 101: 71—82.

Fürst, M. 1978. The effect of acidification on populations of *Astacus astacus*. *In* The first Scandinavian symposium on freshwater crayfish. 1971. Ed.: M. Fürst. Inform. Inst. Freshw. Res., Drottningholm 14: 90—94. (In Swedish with English summary.)

Greenaway, P. 1974. Calcium balance at the postmoult stage of the freshwater crayfish *Austropotamobius pallipes* (Lereboullet). J. exp. Biol. 61: 35—45.

— 1979. Freshwater invertebrates. p. 117—173. *In* Comparative physiology of osmoregulation in animals. Vol 1. Ed.: G. M. O. Maloiy. Academic Press, London.

Haya, K. and B. A. Waiwood. 1981. Acid pH and chorionase activity of Atlantic salmon (*Salmo* *salar*) eggs. Bull. Environm. Contam. Toxicol. 27: 7—12.

JOHANSSON, N., P. RUNN and G. MILBRINK. 1977. Early development of three salmon species in acidified water. Zoon 5: 127—132.

LEIVESTAD, H., G. HENDREY, I. P. MUNIZ and E. SNEKVIK. 1976. Effect of acid precipitation on freshwater organisms. p. 86—111. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

MACKEVICIENE, G. 1975. Studies of the ionic, protein concentration and the protease activity changes in the juveniles of crayfish *Astacus astacus* L. p. 187—194. *In* Freshwater Crayfish II. Pap. Second Int. Symp Freshw. Crayfish, Baton Rouge, La, USA. Ed.: J. W. Awault. Lousiana, State University, Baton Rouge, La.

MALLEY, D. F. 1980. Decreased survival and calcium uptake by the crayfish *Orconectes virilis* in low pH. Can. J. Fish. Aquat. Sci. 37: 364—372.

McDONALD, D. G. and C. M. WOOD. 1981. Branchial and renal acid and ion fluxes in the rainbow trout, *Salmo gairdneri*, at low pH. J. exp. Biol. 93: 101—118.

McMAHON, B. R. and D. O. MORGAN. 1983. Acid toxicity and physiological responses to sub-lethal acid exposures in crayfish. p. 71—85. *In* Freshwater Crayfish V. Pap. Fifth Int. Symp. Freshw. Crayfish, Davis, Cal., USA 1981. Ed.: C. R. Goldman. AVI Publishing Company, Westport, Conn.

McWILLIAMS, P. G. 1980. Acclimation to an acid medium in the brown trout *Salmo trutta*. J. exp. Biol. 88: 269—280.

— 1982. The effects of calcium on sodium fluxes in the brown trout *Salmo trutta*, in neutral and acid water. J. exp. Biol. 96: 439—442.

— 1983. An investigation of the loss of bound calcium from the gills of the brown trout, *Salmo trutta*, in acid media. Comp. Biochem. Physiol. 74A: 107—116.

MORGAN, D. O. and B. R. McMAHON. 1982. Acid tolerance and effects of sublethal acid exposure on iono-regulation and acid-base status in two crayfish *Procambarus clarkii* and *Orconectes rusticus*. J. exp. Biol. 97: 241—252.

NELSON, J. A. 1982. Physiological observations on developing rainbow trout, *Salmo gairdneri*, (RICHARDSON), exposed to low pH and varied calcium ion concentrations. J. Fish Biol. 20: 359—372.

NEVILLE, C. M. 1979. Ventilatory response of rainbow trout *Salmo gairdneri* to increased H^+ concentration in blood and water. Comp. Biochem. Physiol. 63A: 373—376.

PANDIAN, T. J. 1970. Ecophysiological studies on the developing eggs and embryos of the European lobster *Homarus gammarus*. Mar. Biol. 5: 154—167.

PASSANO, L. M. 1960. Moulting and its control. p. 473—536. *In* The physiology of crustacea. Vol 1. Metabolism and growth. Ed.: T. H. Waterman. Academic Press, London.

PETERSON, R. H., P. G. DAYE and J. L. METCALFE. 1980. Inhibition of Atlantic salmon (*Salmo salar*) hatching at low pH. Can. J. Fish Aquat. Sci. 37: 770—774.

ROSSELAND, B. O. 1980. Physiological responses to acid stress in fish. 2. Effects of acid water on metabolism and gill ventilation in brown trout *Salmo trutta* L. and brook trout *Salvelinus fontinalis* MITCHILL. p. 348—349. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

RUNN, P., N. JOHANSSON and G. MILBRINK. 1977. Some effect of low pH on the hatchability of eggs of perch, *Perca fluviatilis* L. Zoon 5: 115—125.

— and M. SOTHELL. 1982a. Sodium influx and transepithelial potential in brown trout (*Salmo trutta* L.) embryos and yolk sac fry at neutral and low pH. *In* P. Runn; Effects of low pH in the development of the early stages of fish. Ph. D. Thesis, Acta Univ. Upsaliensis 647. 20 p.

— and M. SOTHELL. 1982b. Sodium and potassium accumulation in brown trout (*Salmo trutta* L.) embryos and yolk sac fry incubated at neutral and low pH. *In* P. Runn: Effects of low pH on the development of the early stages of fish. Ph. D. Thesis, Acta Univ. Upsaliensis 647. 25 p.

SAS INSTITUTE INC. 1982. SAS User's Guide: Statistics, 1982 Edition. Cary, N. C. SAS Institute Inc., 1982. 584 p.

SHAW, J. 1950. The absorption of sodium ions by the crayfish *Astacus pallipes* LEREBOULLET. III. The effect of other cations in the external solution. J. exp. Biol. 37: 548—556.

STEIN JR, R. A. 1975. Selective predation, optimal foraging and resource depression within the predator-prey interaction between fish and crayfish. Ph. D. Thesis, Univ. Wisconsin, USA. 194 p.

SVÄRDSON, G. 1974. Översikt över laboratoriets verksamhet med plan för år 1974. Inform. Inst. Freshw. Res., Drottningholm (1). 26 p. (In Swedish.)

TROJNAR, J. R. 1977. Egghatchability and tolerance of brook trout (*Salvelinus fontinalis*) fry at low pH. J. Fish. Res. Bd. Can. 34: 574—579.

The Mapping of Short-Term Acidification with the Help of Biological pH Indicators

EVA ENGBLOM and PÄR-ERIK LINGDELL

Myggdalsvägen 120, S-135 43 Tyresö, Sweden

ABSTRACT

We have examined the acidification situation in the Swedish mountain ranges with the help of empirically and experimentally determined tolerance limits for aquatic organisms.

The results show that there is damage due to acidification in the Swedish mountain area. In the Fulufjäll nature reserve in the southern region the benthic fauna has been dominated for the past 10 years by acid-tolerant species such as *Leptophlebia vespertina*, *L. marginata*, *Ameletus inopinatus*, *Baetis rhodani* and *Ephemerella aurivillii* (Ephemeroptera), and *Nemoura cinerea* (Plecoptera). The absence of sensitive species indicates that the pH in streams has often been below 5.0. In the Lake Torrön area in the central mountain region, the acid-sensitive species *Baetis lapponicus* (Ephemeroptera) and *Philopotamus montanus* (Trichoptera), which had been common in 1971, were absent in 1983. The species composition in 1971 indicates that the pH had not previously dropped below 5.5. Judging by the changes in the species composition of the benthic fauna after 1971, however, the pH has been well below 5.0 in many streams. In the Vindelfjäll nature reserve in the northern region those species present in 1961—66 were still represented in 1983 and were dominated by *Baetis lapponicus*. The high frequency of sensitive species indicates that the pH in streams has normally been above 6.0.

I. INTRODUCTION

It is a well-documented fact that thousands of Swedish lakes have been damaged by acidification. Research undertaken in southern Swedish running waters shows that low pH-values and biological damage have also occurred there. The acidification situation in running waters in northern Sweden is not so well known (JOHANSSON and NYBERG 1981, NATIONAL SWEDISH ENVIRONMENT PROTECTION BOARD 1981, 1982, SWEDISH MINISTRY OF AGRICULTURE 1982).

Recorded pH values in running water within the mountain area have usually been above 6.0. This, however, does not necessarily mean that streams here have not on some occasion been acid. In the mountains the pH values of precipitation may be low (Figs. 1 and 2). The pH in the streams decreases as a result of the rapid thawing of large amounts of acid snow.

The magnitude and duration of the drop in pH is dictated by, amongst other things, the character of the catchment area, the pH of the snow, the original pH of the stream, the duration of the spring flood and the increase of flow (DICKSON 1983). Fig. 3 shows the effects of various pH values in the Syterbäcken stream in Sweden's northern mountain region as recorded over two years, one with clean snow and one with polluted snow. When the pH of the snow was 5.0—5.2 (1980) the pH in the stream decreased from 7.55 to 6.25. When the pH of the snow was 4.4 (1979) the pH in the stream decreased from just above 7 to 4.9. At that time the alkalinity was 0.4 mekv/l before the spring flood (FISK and GYDEMO 1979, BJÄRNBORG 1983). In the Delsbo region, a forested area in central Sweden, it was stated that a summer pH of at least 6.5 in streams was necessary to avoid biological damage throughout the rest of the year (ANDERSSON and ANDERSSON 1984). In the Lofsen area in Sweden's southern mountain region, where the pH was normally over 7 and the alkalinity was above 1 mekv/l, the pH decreased to 4.5 in the course of only a few days during the spring flood of 1979. The pH of the snow was then 4.3. Severe damage to the stream's biology was recorded after this pH shock (Olofsson unpubl.).

Bearing in mind the short duration of any given low pH, what essentially is needed at present is continuous measurement in order to ascertain the true acidification situation. One way to get around the problem is to use the tolerance limits of various species to low pH values as a basis for determining the lowest pH attained in streams. Toler-

Fig. 1. The map shows areas investigated and the locations where snow samples were taken.

ance limit is taken as meaning that pH value at which a species has died in aquaria experiments or the lowest pH at which the species has been recorded in nature. As the species composition of the benthic fauna has been shown to relate to the lowest pH value recorded in a stream it is obvious that one single "biological sampling endeavour" can replace several repeated chemical and physical measurements (ENGBLOM and LINGDELL 1983).

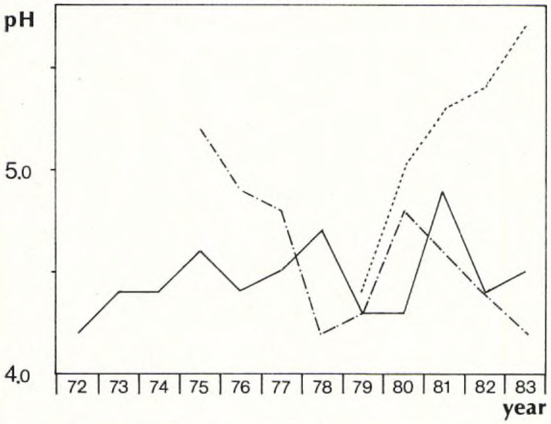


Fig. 2. The pH values of snow from:
——— = The Fulufjäll nature reserve
— · — = The Lofsen area
- - - - - = The Syterbäcken catchment area.

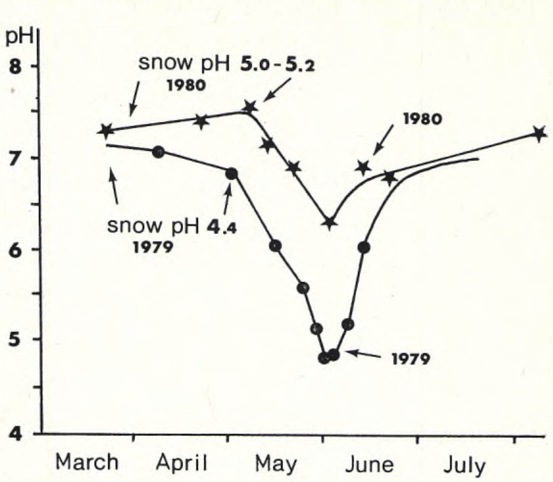


Fig. 3. The effects of pure and polluted snow on the pH of the Syterbäcken stream from 1979—80 (FISK and GYDEMO 1979, BJÄRNBORG 1983).

In 1980, the distribution of various mayfly species (Ephemeroptera) was used as a basis for the mapping of the extent of acidification in small forest streams in Sweden (NATIONAL SWEDISH ENVIRONMENT PROTECTION BOARD 1982). As a follow up to this type of research, the Environment Protection Board began the project "An inventory of the Ephemeroptera of Sweden". One of the aims was to map the short-term pH shocks occurring in the running waters of the Swedish mountain range by means of the changes recorded in the species composition of the benthic fauna.

II. INVESTIGATION AREAS

The investigation areas are presented in Fig. 1. The elevation above sea level ranges from about 400 to over 1,000 m.

The Fulufjäll nature reserve in the southern mountain range is situated mainly on slowly weathering and lime-poor bedrock. Ground cover is thin or absent. The streams within the nature reserve discharge partly into the undammed River Ljördalsälven on the Norwegian side and partly into the undammed River Fuluälven on the Swedish side. Since 1962 areas of the reserve have been the object of liming experiments.

The Lake Torrön area in the central mountain region, like the Fulufjäll nature reserve, is situated on lime-poor and slowly weathering bedrock. Ground cover is very thin or totally absent. The streams drain partly into the undammed Lake Holdern and partly into the regulated Lake Torrön.

The Vindelfjäll nature reserve in the northern mountain range lies in an area of lime-rich bedrock, which is more easily weathered and thus more resistant to acidification. The ground cover is somewhat thicker than in the two other areas. The streams run into the unregulated River Vindelälven.

III. MATERIAL AND METHODS

Benthic fauna from streams within or connected to the Fulufjäll nature reserve have been collected regularly since 1973. In 1983 we visited 17 localities. Analyses of snow have been carried out by the Environment Protection Board since 1972.

Benthic fauna from streams in the Lake Torrön area were collected in 1971, 1974, 1975, 1977 and 1980. We visited 24 localities in 1983.

In the Vindelfjäll nature reserve benthic fauna from streams within or connected to the reserve were collected from 1961—66 (ULFSTRAND 1968). In 1983 we visited 29 localities.

Benthic fauna collected from the three areas in 1983 have been compared with material collected on earlier occasions.

Benthic fauna were collected using a metal cloth dip-net with a diameter of 16 cm and a mesh size of 1.2 mm. At every site at least 30 dip-net samples were taken, covering a botton area of roughly 6 m². On stone bottoms the kick-method was employed. Bottoms with attached vegetation were sampled with a hand net, while soft bottoms were dug up and sieved. All animals found were preserved in 70 % alcohol. All mayflies as well as various other animals which we found interesting from the pH point of view were determined to species. At every locality, the pH was determined in the field.

For the determination of the lowest pH attained in any given stream we have assumed that the pH has not fallen below the tolerance limit for the most sensitive species found in the bottom samples. We have further supposed that if any species typical for a given biotope is absent, and if this species is more sensitive than the most sensitive species actually recorded, then the pH has in all likelihood fallen below the tolerance limit of the missing species. If older material from a localitiy has been available, we have compared this with the 1983 material. When a species which is more sensitive to low pH values than the most sensitive species recorded in 1983 has not been found again 1983, we have assumed that the pH has fallen under the tolerance limit of the missing species.

The reliability of benthic fauna as a pH indicator

To get an idea of how reliable the pH tolerance limit of a given mayfly species is, we compared our results with those of RADDUM and FJELLHEIM (1984) and OTTO and SVENSSON (1983). As shown in Fig. 4, the pH tolerance limits we found by empirical and experimental means for the respective species are often somewhat lower than those empirically obtained in the other studies. These differences may probably be explained by the fact that our background material is decidedly greater and therefore the chances of finding a given species at a lower pH are greatly enhanced. An exception is made for species in which the eggs overwinter, such as *Ephemerella ignita.* Our finds of *E. ignita* came mainly from central and northern Sweden during July—August when pH values are normally high. The pH tolerance limit of 5.0 given by OTTO and SVENSSON (*op.cit.*) is therefore more reliable than our value of 5.9.

Despite the differences in geology and water chemistry between our study and the other studies, the similarities in the recorded tolerance limits for the various species are great. RADDUM and FJELLHEIM (1984) also found *Baetis lapponicus* and *B. macani* to belong to the most sensitive mayflies. They found the pH tolerance limit for both species to be 6.0. Our tolerance limits for these species lie in the range 5.9—6.1. Since we have judged *B. lapponicus* to be the most important indicator animal in the northern mountain range we have presented a sketch of this species (Fig. 5). *B. lapponicus* is the only species of the Nordic mayflies to have an almost totally reduced

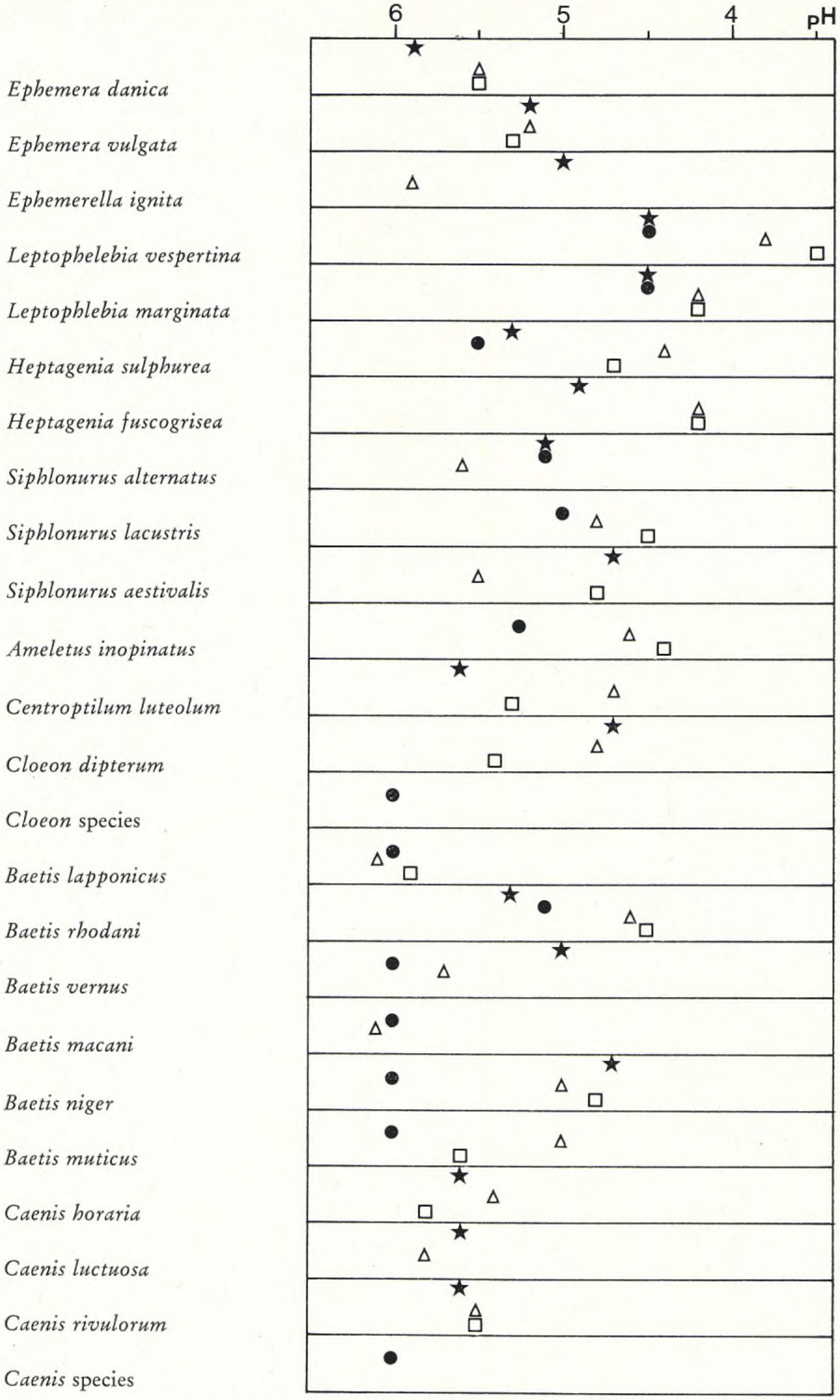


Fig. 4. A comparison of the pH tolerance limits found for different mayflies.

★=OTTO and SVENSSON (1938)
●=RADDUM and FJELLHEIM (1984)
△=Our empirical data
□=Our experimental data.

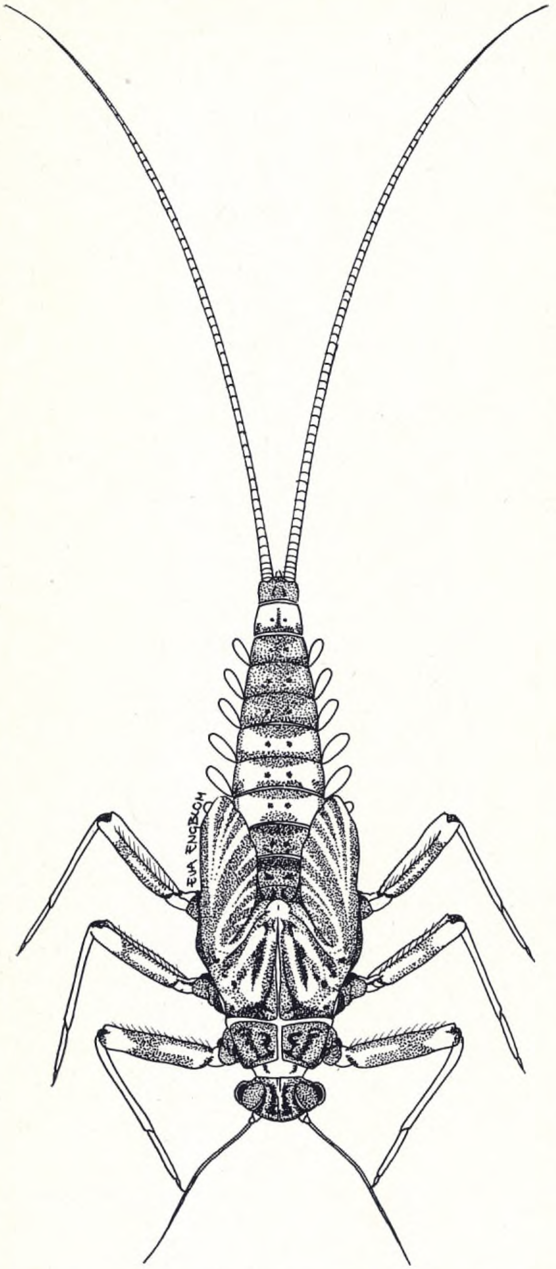


Fig. 5. The mayfly *Baetis lapponicus* is one of those species which has disappeared from many streams in the Swedish mountain chain, due to acidification.

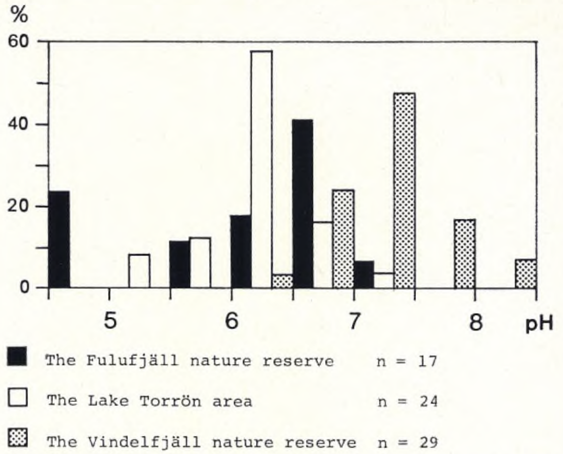


Fig. 6. The percentage of localities in the investigated areas with pH values within the given intervals in 1983.

terminal filament as full-grown larvae. In accordance with our findings, Otto and Svensson (1983) have also found that *Ephemera* and *Caenis* species with a tolerance limit of 5.2 and higher are among the most sensitive mayflies. Raddum and Fjellheim (*op.cit.*) also found the *Caenis* group to be sensitive. The differences between their tolerance limits and ours for these groups do not exceed 0.4 pH units for any species. As in their study, we found the *Leptophlebia* species, which survive pH values of under 4.5 to be the mayflies with the greatest tolerance to acidification.

The pH tolerance limits of 6.0 for *Gammarus lacustris* (Amphipoda), 5.6 for *Dinocras cephalotes* (Plecoptera) and 6.0 for *Philopotamus montanus* (Trichoptera) which Raddum and Fjellheim (1984) found, show good agreement with those found by us (5.5—6.0).

By and large the tolerance limits found by Otto and Svensson (1983) and Raddum and Fjellheim (1984) agree well with those found by us with regard to the species most sensitive to acidification and therefore suitable as pH indicator organisms.

IV. RESULTS AND DISCUSSION

The distribution of recorded pH values within the research areas sampled in 1983 is presented in Fig. 6. The species composition of the bottom samples is presented in Fig. 7. Since the Lake Torrön area revealed the greatest changes in the species composition of benthic animals, these are presented more fully (Table 1 and Fig. 8). The

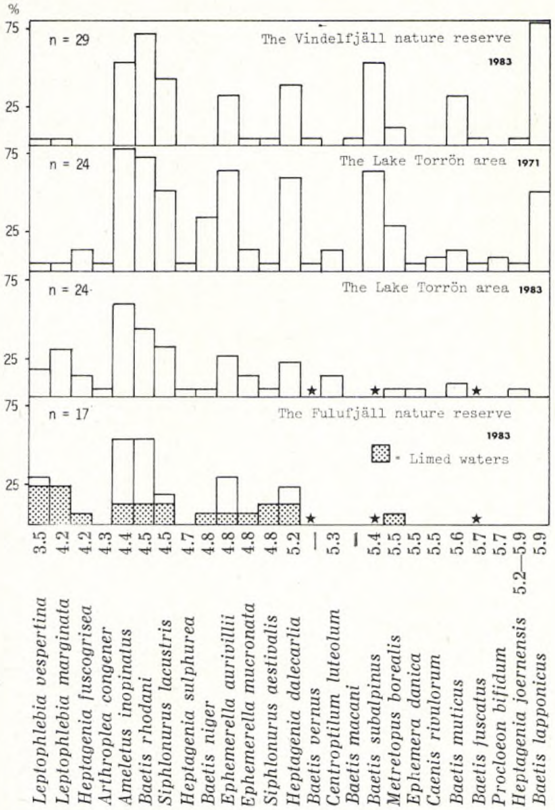


Fig. 7. The percentage of localities in the investigated areas where the different species were present. The figure following the species' name indicates the experimentally determined pH-tolerance limit. An asterisk indicates that the species may have been extremely small or present as eggs on the sampling occasion, which may explain why it was not found.

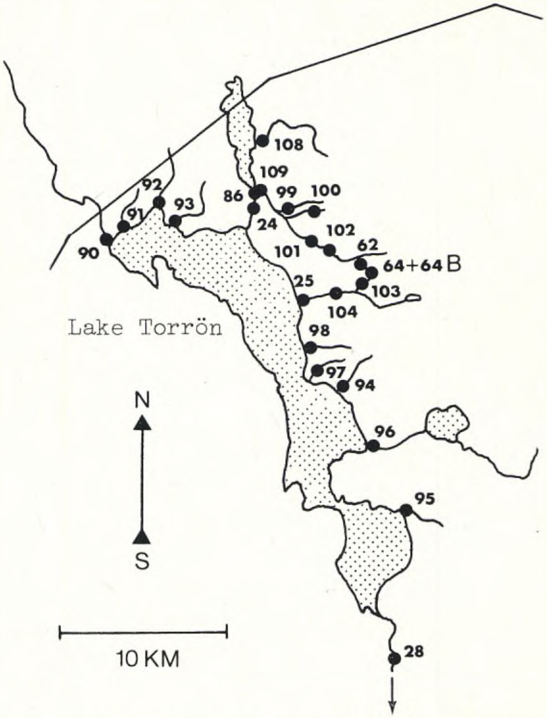


Fig. 8. The map shows the localities in the Lake Torrön area which were investigated in 1971 and revisited in 1983.

experimentally obtained pH tolerance limit for each species is given within parentheses.

Fulufjäll nature reserve: With the exception of a few smaller streams, where the bottom samples are now dominated by extremely acid-tolerant species such as *Nemoura cinerea* (pH 3.5) and the mayfly *Leptophlebia vespertina* (pH 3.5), the fauna in the bottom samples from unlimed water has since 1973 been dominated by the moderately tolerant mayflies *Ameletus inopinatus* (pH 4.4), *Baetis rhodani* (pH 4.5) and *Ephemerella aurivillii* (pH 4.8). More sensitive species such as *Ephemera danica* (pH 5.5) and *Baetis subalpinus* (pH 5.4) have been temporarily recorded during autumn. We have never encountered them after the spring flood, which indicates that the pH has then fallen below their tolerance limits. The species composition from 1973—83 indicates that the pH in unlimed waters has regularly fallen below 5.0. The extremely acid-sensitive mayfly *Baetis macani* (pH 5.5—6.0) has been encountered in limed lakes. In brooks directly downstream of limed lakes the moderately sensitive species *Siphlonurus aestivalis* (pH 4.8) occurred. At a greater distance from the limed lakes, as well as in unlimed streams at higher altitudes, the stonefly *Nemoura cinerea* (pH 3.5) dominated.

Lake Torrön area: During the 1983 inventory, the extremely acid-sensitive species *Baetis lapponicus* (pH 5.9), *B. subalpinus* (pH 5.4) and *Philopotamus montanus* (pH 5.5—6.0), which were common in 1971, were not recorded. That we did not encounter *B. subalpinus* (pH 5.4) is probably due to the fact that at the time of the 1983 visit this species was extremely small or present as eggs. The dominant species in the benthic samples from

Table 1. *Species of mayflies found within the Lake Torrön area in 1971 and 1983. Numbers refer to localities on the map in Fig. 8. For each locality obtained pH and tolerance limits of the most sensitive species found in the respective years are shown.*

+=Found only in 1971 ○=Found only in 1983 *=Found both in 1971 and 1983.

Localities (Jä) [1]		24	25	28	62	64	64B	86	90	91	92	93	94	95	96	97	98	99	100	101	102	103	104	108	109
Obtained pH in 1983		6.9	6.5	7.2	6.2	6.2	6.1	6.9	6.7	5.9	6.0	5.3	6.3	6.6	6.4	5.5	6.1	6.4	6.3	6.2	6.0	6.3	6.3	6.5	6.1
Most sensitive species in 1971. pH limit		5.7	5.9	6.0	5.9	5.9	5.9	5.7	5.2	5.2	4.8	5.5	5.9	5.9	5.6	4.8	4.8	5.9	5.9	5.9	5.9	5.9	5.9	5.5	5.9
Most sensitive species in 1983. pH limit		5.3	5.2	6.0	4.2	4.4	4.2	5.3	5.2	4.4	4.5	4.2	4.8	5.6	5.6	4.2	4.2	4.4	4.5	4.4	4.5	4.5	5.2	4.5	4.5
Leptophlebia vespertina	3.5	*	–	–	○	○	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	○	–	–
Leptophlebia marginata	4.2	○	–	–	○	○	–	*	–	–	○	–	–	–	–	–	○	–	–	○	–	–	–	–	–
Heptagenia fuscogrisea	4.2	*	–	–	–	–	–	*	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	*	–
Arthroplea congener	4.3	*	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Ameletus inopinatus	4.4	*	*	–	+	*	–	–	*	*	*	+	*	*	–	+	+	*	*	*	+	*	*	–	*
Baetis rhodani	4.5	*	*	–	+	+	–	–	*	–	–	–	*	*	*	+	+	+	*	+	+	*	*	–	*
Siphlonurus lacustris	4.5	*	–	*	+	+	–	*	–	–	*	–	–	–	–	–	–	+	–	+	*	*	*	*	–
Heptagenia sulphurea	4.7	–	–	–	–	–	–	–	–	–	–	–	–	–	*	–	–	–	–	–	–	–	–	–	–
Baetis niger	4.8	*	–	–	–	–	–	–	–	+	+	+	–	–	–	–	–	+	+	+	–	+	–	–	–
Ephemerella aurivillii	4.8	*	*	–	–	–	–	–	*	+	+	+	*	–	*	+	+	+	+	+	–	+	*	–	–
Ephemerella mucronata	4.8	*	–	–	–	–	–	*	*	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Siphlonurus aestivalis	4.8	+	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Heptagenia dalecarlia	5.2	*	*	–	+	+	–	–	*	+	–	–	–	*	–	–	–	+	+	+	+	+	*	–	+
Baetis subalpinus	5.4	+	+	–	+	+	–	+	–	–	–	–	–	+	+	–	–	+	+	+	+	+	+	+	+
Baetis vernus	—	+	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Centroptilum luteolum	5.3	*	–	–	–	–	–	*	–	–	–	–	–	–	*	–	–	–	–	–	–	–	–	–	–
Metretopus borealis	5.5	+	–	*	–	–	–	+	–	–	–	+	–	–	–	–	–	+	–	–	–	–	–	+	+
Ephemera danica	5.5	–	–	*	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Caenis rivulorum	5.5	+	–	+	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Baetis muticus	5.6	+	–	–	–	–	–	–	–	–	–	–	–	*	*	–	–	–	–	–	–	–	–	–	–
Baetis fuscatus	5.7	+	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Procloeon bifidum	5.7	+	–	+	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Heptagenia joernensis	5.2—5.9	–	–	*	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–	–
Baetis lapponicus	5.9	–	+	–	+	+	+	–	–	–	–	–	+	+	–	–	–	+	+	+	+	+	+	–	+

---

[1] Refers to the province of Jämtland.

1983, as in the Fulufjäll nature reserve from 1973—83, were the moderately acid-tolerant mayflies *Ameletus inopinatus* (pH 4.4), *Baetis rhodani* (pH 4.5) and *Ephemerella aurivillii* (pH 4.8). An increase in numbers of the extremely tolerant species *Leptophlebia vespertina* (pH 3.5) and *L. marginata* (pH 4.2) was observed in streams which, according to the local inhabitants, had earlier yielded the now absent brown trout (*Salmo trutta*). The important difference in the species composition between 1971 and 1983 is that those species which had empirically and experimentally been shown to be sensitive to low pH values were also those which were not encountered in 1983 (Fig. 7 and Table 1). Fig. 9 presents a comparison of the distribution of the pH values recorded in 1983 and the pH tolerance limits for the most sensitive species recorded in 1971 and 1983. As is shown in Fig. 9 the bottom samples from streams in 1971 were dominated by species with a tolerance limit of pH 5.5—6.0. In 1983 the bottom samples from streams were dominated by species with a tolerance limit of pH 4.0—4.5. This indicates that the lowest pH in the majority of streams at some point between 1971 and 1983 fell two pH units below the 1983 value. It is logical to assume that the pH of the snow in 1979, as in the areas south and north of the Lake Torrön area, was acid (Figs. 1 and 2), and that the pH in the weakly buffered streams fell below 5.0 (Fig. 3). A comparison of the species composition in bottom samples from the Fulufjäll nature reserve in 1983, where the pH of the snow has fallen below 5.0 since 1972, and the Lake Torrön area in 1983 shows that these are in general identical. A comparison of bottom samples from the Lake Torrön area in 1971 and the Vindelfjäll nature reserve in 1983, where the pH of the snow has exceeded 5.0 from 1980 onwards, also shows great similarities.

Vindelfjäll nature reserve: Judging from the bottom samples, *Baetis lapponicus* (pH 5.9) was the most wide-spread species in the area in 1983. None of the localities visited by Ulfstrand from 1961—66 lacked *B. lapponicus*. The species composition in bottom samples from localities investigated by Ulfstrand (1968) and revisited by us in 1983 were in general the same for comparable biotopes. This indicates that the pH must not have fallen below 6.0. neither in 1961—66 nor in 1983.

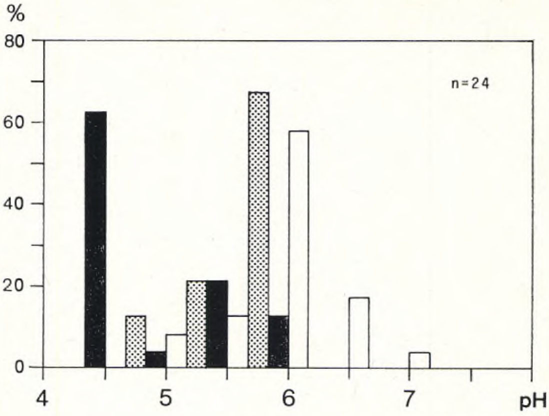


Fig. 9. The percentage of localities in the Lake Torrön area with pH values within the given intervals, and the percentage of localities where the tolerance limits of the most sensitive species fell within the same pH intervals in 1971 and 1983.

□ = pH value measured in 1983
▨ = pH tolerance limit of most sensitive species found in 1971
■ = pH tolerance limit of most sensitive species found in 1983.

The bottom samples from streams in the Vindelfjäll nature reserve were generally richer in both species and individuals than the bottom samples from streams in the Fulufjäll nature reserve. The important difference was that the bottom samples from streams in the Vindelfjäll nature reserve showed a high frequency of those species which had been shown empirically and experimentally to be sensitive to low pH values. The species composition in bottom samples from the Vindelfjäll nature reserve were greatly similar to those from the Lake Torrön area in 1971.

V. CONCLUSIONS

Fulufjäll nature reserve: The pH in larger unlimed streams often falls below 5.0. These streams are dominated by the tolerant mayflies *Baetis rhodani* and *Ameletus inopinatus*, both of which can tolerate pH values down to 4.5. Smaller unlimed streams are permanently acidified today and are dominated by the stonefly *Nemoura cinerea* which tolerates pH values down to 3.5. Liming has led to acceptable conditions in lakes for the sensitive mayfly *Baetis macani* which tolerates pH values down to 5.5.

Lake Torrön area: Judging by the changes in the species composition of the bottom samples here, the pH situation has changed drastically since 1971. The species composition of 1971 indicates that pH values lower than 6.0 were unusual in streams. In 1983 the situation is the opposite. The mayfly *Baetis lapponicus* and the caddisfly *Philopotamus montanus*, which were common in 1971, were not encountered at all in 1983. In 1983 the area was dominated by the mayfly *Ameletus inopinatus* which tolerates pH values down to 4.5. This indicates that several streams have had pH values well below 5.0 between 1971 and 1983. The number of permanently acidified waters where *Leptophlebia marginata* dominates has increased.

Vindelfjäll nature reserve: The high frequency of sensitive species in 1961—66 as well as in 1983 indicates that the pH during these years has probably not fallen below 6.0 in the streams of the reserve. They were dominated in 1983 by the sensitive mayfly *Baetis lapponicus* which is damaged by pH values just under 6.0.

VI. REFERENCES

ANDERSSON, B. and P. ANDERSSON. 1984. The distribution of trout (*Salmo trutta*) relation to pH — an inventory of small streams in Delsbo, central Sweden. Rep. Inst. Freshw. Res., Drottningholm 61: 28—33.

BJÄRNBORG, B. 1983. Dilution of and acidification effects during the spring flood of four Swedish mountain brooks. Hydrobiologia 101: 19—26.

DICKSON, W. 1983. Characterization of acidified waters. VDI-Berichte 500: 387—391.

ENGBLOM, E. and P.-E. LINGDELL. 1983. Bottenfaunans användbarhet som pH-indikator. Rep. Nat. Swedish Environm. Protec. Bd. SNV PM 1741. 181 p. (In Swedish with English abstract.)

FISK, E. and R. GYDEMO. 1979. pH-mätning i snö och smältvatten i Västerbottens läns fjälltrakter våren 1979. Fiskenämnden och Lantbruksnämnden i Västerbottens län, Umeå, Sweden. 17 p. (Mimeographed in Swedish.)

JOHANSSON, K. and P. NYBERG. 1981. Acidification of surface waters in Sweden — effects and extent 1980. Inform. Inst. Freshw. Res., Drottningholm (6). 118 p. (In Swedish with English summary.)

NATIONAL SWEDISH BOARD OF FISHERIES and NATIONAL SWEDISH ENVIRONMENT PROTECTION BOARD. 1981. Liming of lakes and rivers 1977—1981 in Sweden. Inform. Inst. Freshw. Res., Drottningholm (4). 201 p. (In Swedish with English summary.)

NATIONAL SWEDISH ENVIRONMENT PROTECTION BOARD. 1981. Monitor 1981 — försurning av mark och ytvatten. Nat. Swedish Environm. Protec. Bd. Medd. 3. 175 p.

— 1982. Ecological effects of acid deposition. Rep. 1982 Stockholm Conf. Acid. Environm. Rep. Nat. Swedish Environm. Protec. Bd SNV PM 1636. 340 p.

OTTO, C. and B. SVENSSON. 1983. Properties of acid brown water streams in South Sweden. Arch. Hydrobiol. 99: 15—36.

RADDUM, G. G. and A. FJELLHEIM. 1984. Acidification and early warning organisms in freshwater in Western Norway. Verh. intern. Verein. Limnol. 22. (In the press.)

SWEDISH MINISTRY OF AGRICULTURE. 1982. Acidification today and tomorrow. The 1982 Stockholm conference on the acidification of the environment. 231 p.

ULFSTRAND, S. 1968. Benthic animal communities in Lapland streams. Oikos Suppl. 10. 120 p.

Aluminium Toxicity to Atlantic Salmon (*Salmo salar* L.) and Brown Trout (*Salmo trutta* L.): Mortality and Physiological Response

SVEINUNG FIVELSTAD and HELGE LEIVESTAD

University of Bergen, Zoological Laboratory, N-5000 Bergen, Norway

ABSTRACT

Mortality and several physiological stress parameters have been registered for different life stages of Atlantic salmon and brown trout, exposed to varying levels of aluminium in an open flow system.

Salmon swim-up larvae were more sensitive than postlarvae when exposed to Al-concentrations from 110 to 300 µg/l at pH 5, both naturally occurring in acid water supply, and added as $Al_2(SO_4)_3$. Survival time (L_{50}) decreased with increasing Al-concentration and the dose-response relationship was better correlated with the $Al(OH)^{2+}$ monomer than with Al^{+++}, $Al(OH)_2^+$ or AlF^{2+}.

Exposure of salmon parr to natural variation in Al (50—180 µg/l) at pH 5.3 induced a hyperventilatory response that was maintained for 26 days, when compared with parr exposed to pH 5.5. Hyper-ventilation was induced in pH 5.5 when the Al level was raised to 250 µg/l at day 27. Increased hematocrit and minor reduction in chloride were observed during this sublethal stress.

Brown trout exposed at pH 5 to the same Al regime showed no sublethal stress symptoms. After 34 days acclimation in this water, trout were exposed to 360 µg/Al (pH 5) and had acquired improved tolerance compared with control. Acclimation had occurred both in respiratory parameters and in salt balance. Mortality was associated with coughing, hyperventilation, increased haematocrit and reduced plasma levels for sodium, chloride and osmolarity.

I. INTRODUCTION

Acid precipitation leach aluminium from the soil resulting in elevated concentrations of aluminium in lakes and rivers in Scandinavia and North America (CRONAN and SCHOFIELD, 1979; DRISCOLL, 1980; DICKSON, 1980). In natural waters aluminium will be complexed with organic ligands and with hydroxide, fluoride and sulfate ions. Aluminium hydroxide complexes also tend to polymerize, particularly at high aluminium concentrations (MAY *et al.* 1979), but in natural acid water the level of polymeric complexes is considered to be low (DRISCOLL *et al.* 1982).

To explain the decline in fish populations the role of aluminium has been considered (MUNIZ and LEIVESTAD, 1980 a; SCHOFIELD and TROJNAR, 1980). Aluminium complexed with organic ligands apparently is not toxic, and aluminium chelated with fluoride seems to be of lower toxicity than the free ion and the monomeric hydroxy complexes (BAKER and SCHOFIELD, 1980). Most fish kills in acidified waters occur in the pH-range 4—6. The inorganic monomeric species highest in concentrations in this pH-interval are Al^{3+}, $Al(OH)^{2+}$, $Al(OH)_2^+$ and AlF^{2+}. The relative concentrations of these species depend on pH and on the total fluoride level. The free ion dominates in concentration at pH 4, and decreases in concentration with higher pH. The maximal concentrations of the hydroxy complexes occur close to pH 5, decreasing at higher and lower pH. Aluminium toxicity has also been shown to be pH-dependent with maximum toxicity around pH 5 (BAKER and SCHOFIELD, 1980; MUNIZ and LEIVESTAD, 1980 b), suggesting that $Al(OH)^{2+}$ and (or) $Al(OH)_2^+$ are the most toxic aluminium species. The mode of action of aluminium is still unknown, but hyperventilation, coughing and loss of salt from the blood plasma have been observed in brown trout (*Salmo trutta* L.) exposed to 150—900 µg/l Al at pH 5 (MUNIZ and LEIVESTAD, 1980 a). In this Al-range the trout died and little is known about the physiological responses under sublethal Al-stress.

Acidified Norwegian rivers have lost their populations of Atlantic salmon (*Salmo salar* L.)

earlier than the brown trout (JENSEN and SNEKVIK, 1972). Bioassays in water from an acid river also suggest that brown trout are less sensitive than Atlantic salmon (GRANDE *et al.* 1978). In these bioassays, the hydrogen ion and aluminium can both have been a toxic stimulus. It is therefore not certain that brown trout has a lower sensitivity to aluminium than Atlantic salmon, but this may possibly be the case.

The present investigations were done to study the potential for aluminium toxicity to Atlantic salmon and brown trout in acid water. The physiological responses at sublethal and lethal Al-stress are considered, and attempts to relate mortality to concentrations of individual Al-species are made.

II. MATERIAL AND METHODS

The swim-up larvae and postlarvae (fry after initiation of feeding) of Atlantic salmon used in trial A and B were obtained from the Institute of Marine Research hatchery at Matre, April 8, 1981. Trial A was started April 11 and trial B May 5. The fish used in trial A was in the swim-up stage, and the postlarvae exposed in trial B had passed this stage about two weeks earlier. The swim-up larvae used in trial C were fetched at Matre on May 15. This trial was started one week later.

The two-year-old Atlantic salmon parr (25—79 g) and the two-year-old brown trout (37—120 g) used in trial D were obtained from the Norwegian Directorate for Wildlife and Freshwaterfish hatchery at Ims on September 9, 1981. The trial started on September 15.

Mortality was used as a measure of toxicity in trial A, B and C. These trials lasted until at least 50 % mortality had occurred. Fish were judged to be dead when opercular movements had ceased and no swimming response could be elicited through stimulation of the caudal peduncle. Mortality was quantified in terms of L_{50}, the time until 50 per cent of the fish are dead. The average number of fish exposed in each water-quality was 161.

In trial D ventilation rate, haematocrit and bloodplasma levels of sodium, chloride and osmolarity were measured. Blood samples were taken by cardiac puncture into 1 ml heparinized syringes and centrifuged for 5 minutes. Haematocrit was measured, and the plasma samples were immediately analysed for osmolarity and chloride. Plasma samples were stored frozen for later sodium analyses. Sodium was determined by flame emission using a Pye Unicam Atomic Absorption Spectrophotometer, on plasma samples diluted 1/200. Plasma chloride concentration was measured coulometrically using a Radiometer CMT 10 Chloride Titrator (10 μl samples). Osmolarity was measured on 8 μl samples with a Wescor 5100G vapor pressure osmometer. Ventilation frequency was measured by visual inspection on 5—10 fish randomly selected.

The tapwater used in the experiments derives from the River Mulelven catchment where only the upper half is impounded by Lake Storediket. The catchment is very steep with thin soil cover and is largely underlain by grey and red micaceous gneiss and migmatite bedrock. The water is soft (Ca < 1 mg/L) and acid, and the water quality changes rapidly after heavy rain. Annually the pH varies between 4.55 and 5.20 and aluminium between 60 and 400 μg/l (Fe: 40—140 μg/l; Mn: 10—17 μg/l; Na: 2.5—3.9 mg/l). The raw water is first adjusted to pH 6.2—6.7 by adding KOH via an automatic pH-control system. Our control fish were kept at this pH-level. Controlled Al/pH-combinations were reached by continuously adding $Al_2(SO_4)_3$ and H_2SO_4 to the neutralized water, using peristaltic pumps.

The fish were kept in keep-nets in 300 litre tanks fed continuously with freshly mixed aluminium, acid and water at a rate of 5 litre/min. The "age" of the water in the tanks should thus be constant throughout the experiments (halflife 30 minutes). During the experiments the tapwater had varying concentration of aluminium. In some of the bioassays the fish were exposed to only this aluminium level.

Assuming thermodynamic equilibrium between Al-species in our test water, the concentration of individual species was calculated. According to SMITH (1971), the activities of individual monomeric species adjust themselves almost instantly with pH. The equilibrium constants used are from BERSILLON *et al.* (1978), MAY *et al.* (1979) and SMITH (1971). The calculations were based on measured values of pH, inorganic Al-level, and total fluoride and sulfate concentration (average: F 34 μg/l; SO_4 3.4 μg/l). The level of Al complexed with organic ligands is usually found to be low in the pH-range used in the mortality trials, about 20 μg/l, and does not depend on the total Al-level (FIVELSTAD, 1983). 20 μg/l was therefore subtracted from the total Al-concentration in trial A and B. In trial C and D inorganic and organic Al-species were separated by a cat-ion exchange column saturated with sodium, as proposed by DRISCOLL *et al.* (1982).

Total Al-concentration was determined at least 3 times per week in each water quality. The Al-concentration was determined spectrophotometrically by the pyrocatechol method (DOUGAN and WILSON 1974). pH was measured and adjusted (if necessary) at least once per day in each water-quality. For the most part, pH-levels were maintained within 0.1 units of average pH-levels.

A lgK-value of 10.18 for the reaction $Al(OH)_3 + 3H^+ = Al^{3+} + 3H_2O$, was used in the calculations. Using ultrafiltration (0.01 μ) as an indicator for existence of precipitate (supersaturation), the ion-exchangeable fraction (monomers) in our water gave lgK-values 10.18 ± 0.09 ($n = 10$). Water samples were immediately filtered and passed directly into the ion-exchange column. The samples were selected from pH 5.4—5.9 at temperatures from 4—6°C. The variations in lgK values indicate that our methods for isolating the different fractions are far from perfect.

III. RESULTS AND DISCUSSIONS

Mortality trials

The Al-concentration in the tapwater used was never below 110 μg/l during the mortality tests. To study possible effects of acid alone, 15 mg/l citrate was added to the water at pH 4.9 and at an Al-level of 140 μg/l. BAKER and SCHOFIELD (1980) have demonstrated that the aluminium-citrate complex is not toxic to fish. Only 1 out of 200 swim-up larvae died in this water quality during 108 hours' exposure, and no behavioural response was seen, indicating that no mortality was caused by acid alone in our trials. At all other pH/Al-combinations used in trial A and B hyperventilation and coughing were observed. The fish were sluggish and had almost no avoidance reaction, and spasms were observed before the fish were dying.

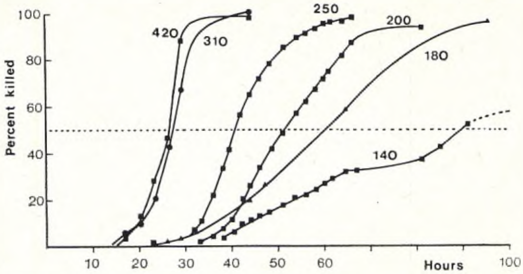


Fig. 1. (Trial A): Accumulated mortality for swim-up larvae of Atlantic salmon exposed to Al at pH near 5 (Table 1), plotted against length of exposure. The numbers refer to mean total Al-concentration in μg/l. At the two lowest Al-levels, Al is only of natural origin. Temperature 6.0°C.

The results from the mortality trials with swim-up larvae are presented in Fig. 1. The experiments were performed at pH levels around 5 (Table 1) and it may be noted that the survival times for 310 and 420 μg/l total Al are virtually identical. This may indicate some sort of "saturation" of the toxic effect. The mortality curve at 140 μg/l has an irregular shape and only 62 % mortality was reached after 128 hours. This may indicate that acclimation may take place at this Al/pH-level.

The results from experiments with older larvae (20 days, trial B) are presented in Fig. 2. It may

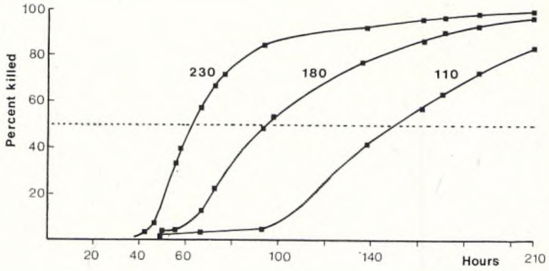


Fig. 2. (Trial B): Accumulated mortality for Atlantic salmon postlarvae exposed to Al at pH near 5 (Table 1), plotted against length of exposure. The postlarvae had passed the swim-up stage about two weeks earlier. The numbers refer to total Al-concentration in μg/l. At the lowest Al-level, Al is only of natural origin. The temperature was 6—7°C until 140 hrs., but had increased to 11°C at the end of the experiment.

Table 1. *Calculated concentrations ($\times 10^{-7}$ M) of the inorganic aluminium species together with L_{50} for two developemental stages of Atlantic salmon. Total inorganic Al ($Al_{inorg.}$), pH, fluoride — and sulfate concentration are used in the calculation.*

	Swim-up larvae (trial A)						Postlarvae (trial B)		
pH	5.16	5.29	4.85	4.89	5.18	5.00	5.11	5.11	5.08
$Al_{inorg.}$	148.2	107.4	85.2	66.7	59.3	44.4	77.8	59.3	33.3
Al^{3+}	31.2	14.9	31.4	21.5	9.9	10.1	16.7	11.8	5.6
$Al(OH)^{2+}$	45.1	29.0	22.2	16.7	15.0	10.1	21.5	15.2	6.8
$Al(OH)_2^+$	51.8	44.9	12.5	10.3	18.0	8.0	22.0	15.6	6.5
$Al(OH)_4^-$	0.9	1.4	0.1	0.1	0.3	0.1	0.3	0.2	0.1
AlF^{2+}	16.8	15.3	16.8	16.2	14.3	14.3	15.6	14.8	12.5
AlF_2^+	0.5	0.9	0.5	0.7	1.1	1.1	0.8	1.1	1.5
$AlSO_4^+$	1.7	0.8	1.7	1.2	0.6	0.6	0.9	0.7	0.3
L_{50} (hrs.)	26	27	41	52	60	89	62	93	150

be noted that even at 110 µg/l the shape of the mortality curve does not indicate effects of acclimation. It must be noted that the animals were not fed during the experiment and that the temperature increased towards the end. Effects from starvation can thus not be excluded.

Toxic Al.

The time for 50 per cent mortality (L_{50}) have been read from Fig. 1 and Fig. 2. Table 1 shows calculated levels of the inorganic monomeric Al-species. The concentrations of $Al(OH)_4^-$, $AlSO_4^+$ and AlF_2^+ are low at all the Al/pH-combinations used, and can presumably be ignored as toxic agents in these trials. The level of AlF^{2+} is almost the same in all water qualities, particularly in trial A. The only species which increase in concentration with decreasing survival time (L_{50}) are Al^{3+}, $Al(OH)^{2+}$ and $Al(OH)_2^+$.

Because Al is most toxic in the pH range 5.0—5.4 (BAKER and SCHOFIELD, 1982; MUNIZ and LEIVESTAD, 1980 a) the Al^{3+}-ion cannot be of high toxicity. Our data from trial C (Fig. 3) suggest the same. Survival time for swim-up larvae was longer at pH 4.7 than at pH 5.0 although the level of Al^{3+} is almost doubled from pH 5.0 to pH 4.7 (Table 2). At pH 4.7 the hydrogen ion may also be toxic.

Mortality (L_{50}) on both swim-up larvae and postlarvae (trial A and B) are significantly correlated with total inorganic Al-level ($p < 0.01$), but since Al-toxicity is pH-dependent, all species cannot be of equal toxicity. As earlier investigations have suggested, therefore, either $Al(OH)^{2+}$ or $Al(OH)_2^+$, or both these species, have the highest toxicity (MUNIZ and LEIVESTAD, 1980 a).

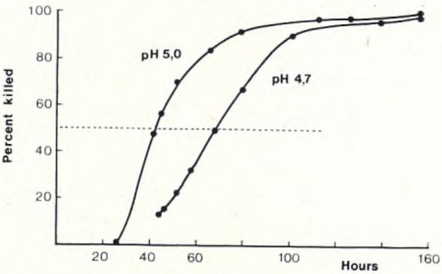


Fig. 3. (Trial C): Accumulated mortality of Atlantic salmon swim-up larvae exposed to 280—300 µg/l Al at pH 4.7 and pH 5.0. Temperature 11.7°C.

Simple linear regression analyses were performed between L_{50} and log-transformed concentrations of Al-species. (Values from trial C were ignored due to higher water temperature in this trial.) Mortality on swim-up larvae (trial A) was significantly correlated with the concentrations of $Al(OH)^{2+}$ ($p < 0.01$), but was not significantly correlated with the level of Al^{3+} ($p < 0.10$) or the level of $Al(OH)_2^+$ ($p > 0.10$). Mortality of postlarvae (trial B) was significantly correlated with both Al^{3+} and the two hydroxy complexes ($p < 0.05$), but this seems to be a result of the experimental conditions used: pH was almost the same in the three water-qualities used in trial B (cf Table 1), keeping the relative relationship between species constant.

The data from the mortality trials thus seem to indicate that $Al(OH)^{2+}$ is the most toxic Al-

Table 2. *Measured concentrations of inorganic Al and calculated concentrations of inorganic Al-species ($\times 10^{-7}$ M). Al chelated with organic ligands was measured to 20 µg/l (7.4×10^{-7} M) at both pH-values.*

pH	inorg. Al	Al^{3+}	$Al(OH)^{2+}$	$Al(OH)_2^{+}$	AlF^{2+}	$Al(SO_4)^{+}$
5.0	88.9	24.4	24.8	20.4	17.4	2.2
4.7	96.3	44.8	21.5	8.2	16.7	3.7

species. The correlation found between $Al(OH)^{2+}$-concentration and mortality can be a matter of coincidence, but it represents the simplest model accounting for our data. More than one species may be toxic, and the different species can have different toxicity. At low Al-concentrations the toxicity of the AlF^{2+} complex will be of particular importance (Table 1), because a high proportion of inorganic Al can exist as AlF^{2+} in the pH-range 4.0—5.3, depending on total fluoride level.

With these considerations in mind, mortality on swim-up larvae and postlarvae has been plotted against the concentration of the $Al(OH)^{2+}$-complex (Fig. 4). The abscissa is logaritmic, giving straight lines for an exponential relationship. Swim-up larvae seem to be more sensitive to Al than postlarvae. The toxic mechanisms causing death on swim-up larvae seem to have reached saturation at a concentration of about 2.9×10^{-6} M. No lower lethal threshold was indicated for swim-up larvae or postlarvae, because of the aluminium-rich tapwater used.

If the $Al(OH)^{2+}$-complex is the main toxic substance in acid waters, the toxicity will have its maximum at pH 5.0, decreasing at higher and lower pH. High levels of fluoride ions and organic ligands can shift the maximum concentration of $Al(OH)^{2+}$ to a higher pH. This is in agreement with the conclusions of BAKER and SCHOFIELD (1982): Aluminium is most toxic in the pH-range 5.2—5.4. The fluoride concentration of their test water was 5—6 times our own.

It has been suggested that supersaturated Al-solutions, *i.e.* when $Al(OH)_3$ starts to precipitate, are toxic to fish. It should be recalled that our "reference water" often has high levels of Al (up to 300 µg/l). The raw water is titrated to pH 6.5 with KOH and fed into the fish tanks only 4 minutes after neutralization. Brown, metallo-humic, precipitate rapidly accumulate in tanks and piping, and nearly 50 % of total Al can be removed by filtering (0.05 µ) freshly prepared reference water. A long row of experiments have been performed in this water over the last 3 years, keeping the most sensitive stages of salmon and other species for months. We have not had a single incidence of mortality or seen any sublethal stress symptoms as long as pH is kept between 6 and 7. We find it highly unlikely, therefore, that $Al(OH)_3$-complexes are toxic to fish.

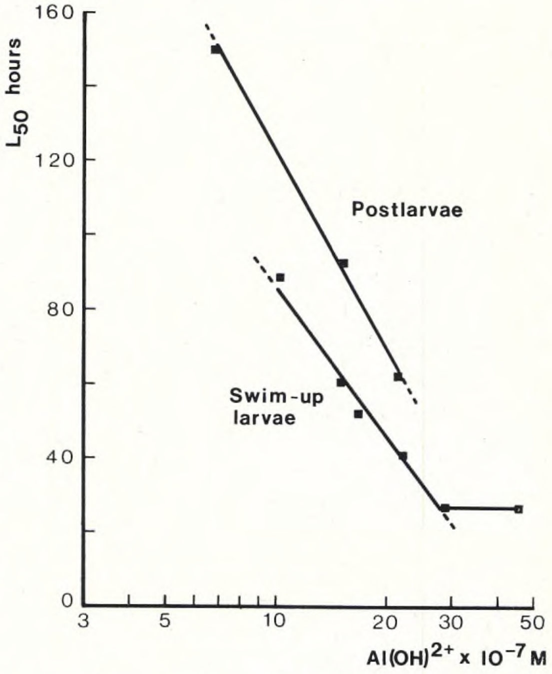


Fig. 4. Mortality for swim-up larvae (Trial A) and postlarvae (Trial B) of Atlantic salmon plotted as a function of calculated concentrations of the $Al(OH)^{2+}$-complex. The letters on the figure refer to the trials, and the lines are regression lines.

Postlarvae: $Y = -1.74 \times 10^5 \log X + 296$, $r = -1.00$.
Swim-up larvae: $Y = -1.33 \times 10^5 \log X + 218$, $r = -0.99$.

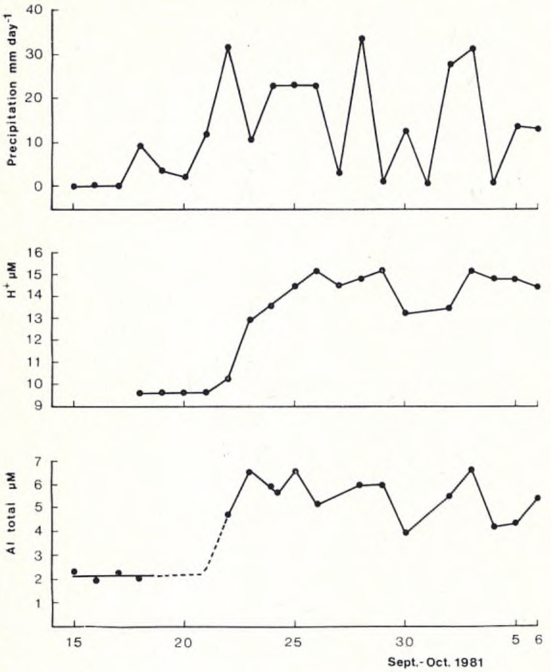


Fig. 5. Temporal variations in precipitation, H⁺- and Al-concentrations, September 15 to October 6, 1981. From September 9 to September 15 no precipitation occurred.

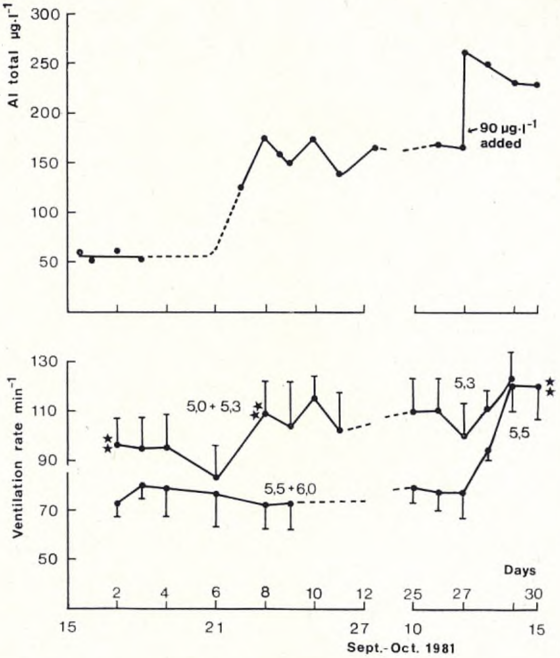


Fig. 6. (Trial D): Ventilation frequency of two-year-old Atlantic salmon parr subjected to variations in Al-concentration at different pH-values (numbers on the figure). ** = significant difference from control group ($p < 0.01$, Wilcoxon rank test). Control values are identical to pH 6.0 values. Means and SD are given (n = 10 for September; n = 5 for October). The temperature fell from 12.8°C, September 15 to 10°C, October 15.

Sublethal stress

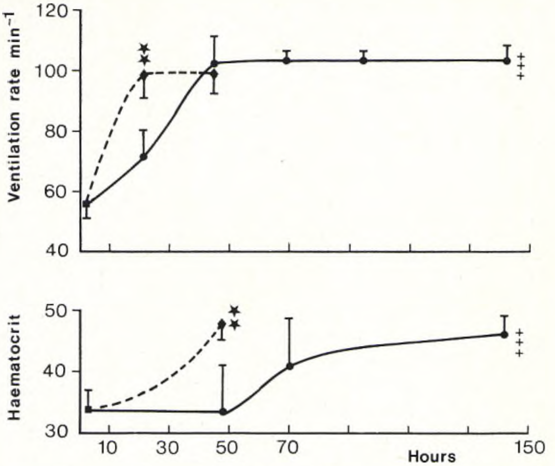
In trial D two-year-old Atlantic salmon parr and brown trout were subjected to four different pH-values and varying Al-levels. The first 27 days the fish were exposed to only naturally occurring Al. The concentration of Al in our tapwater was low after a period with no precipitation September 9—17, but H^+- and Al-concentration increased after rainfall September 20—26 (Fig. 5). Respiratory frequencies for parr in experiment D are presented in Fig. 6. The data from pH 5.5 and 6.0 are grouped from day 2 to 27 since they showed no significant difference in this range of Al-concentration. In our context this frequency is taken as the control level. Similarly, frequencies registered at pH 5.0 and 5.3 are grouped, since differences here also are insignificant.

At pH 5.0—5.3 parr ventilation rate was increased after 2 days' exposure to 50—70 µg/l Al ($p < 0.01$), but had decreased to near control values after 6 days' exposure. The reduction was significant ($p < 0.05$), suggesting partial acclimation at these pH/Al-combinations, but the ventilation rate again increased when Al-concentration increased after rainfall (day 8—11).

From day 12 to day 25 the ventilation rate was not measured. The fish were out of the keep-nets for feeding, but parr exposed to pH 5.3 was observed to hyperventilate during this period. The Al-concentration varied between 110- and 180 µg/l these days (mean value = 145 µg/l).

After 27 days exposure to only naturally occurring Al, 90 µg/l Al was added to the water, giving a total Al-concentration in the range 230—250 µg/l. At this Al-level, the ventilation rate of parr exposed to pH 5.5 also increased.

The data presented in Fig. 6 first of all indicate that measurement of respiratory frequency may be a valuable tool for recording sublethal stress in acid, aluminium-rich water. Rosseland (1980) reported similar hyperventilatory responses in



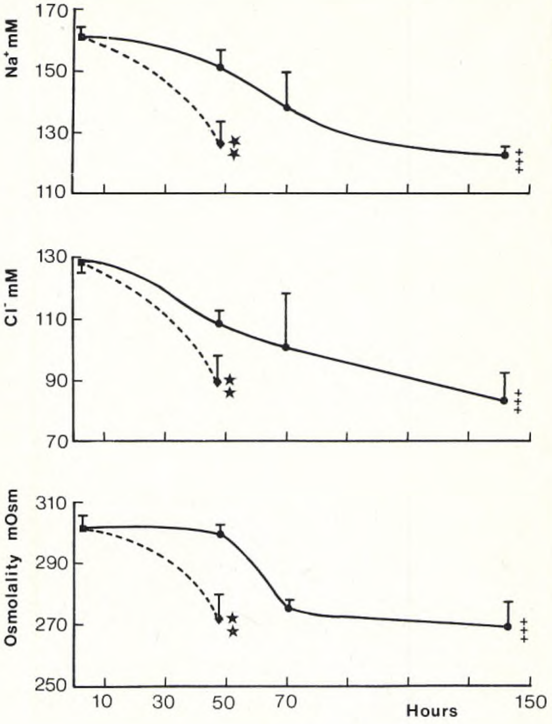


Fig. 7. (Trial D): Physiological response in acclimated (●), 34 days (see text) and non-acclimated (◆) brown trout, subjected to 360 μg/l total-Al, at pH 5.0—5.2 and 8°C. Control: Trout from pH 6.3 (■). **=significant difference between acclimated and non-acclimated. ($p < 0.01$, Wilcoxon rank test). Means and SD are given. (n=10 for acclimated trout, 48 hrs., elsewhere n=5). +=dead.

(a): Ventilation frequency, haematocrit;
(b): Plasma Na^+, Cl^- and osmolarity.

brown trout and brook trout, both at acutely lethal pH/Al-levels and lower. Our experiments lasted for 29 days without mortality, and from day 2 to day 27, smolt kept at pH 5.3 had significantly higher ventilatory frequency than those kept at pH 5.5. It should be borne in mind that the only difference in water quality between the two groups is the amount of sulfuric acid added to keep the pH at the proper level. Both total Al and the ion exchangeable fraction were within the analytical accuracy when comparing tank water from pH 5.3 and 5.5. The hyperventilatory response in the experiment therefore must be initiated and maintained by minute shifts in aluminium speciation or in the H^+-level. It seems futile to speculate over the toxic agent in this experiment, since the fish obviously is a better sensor for chemical differences than our analytical methods.

The difference in response between the two groups, after 90 μg/l Al was added at day 27, may indicate either an acclimation for the 5.3 group, or that near-maximum hyperventilation had already been reached for this group.

Blood samples were taken from the parr in the different pH/Al-combinations. Haematocrit and plasma chloride values are given in Table 3, together with ventilation rate of parr in the same water-quality. Most groups had increased haematocrit, but the increases were not significant for all groups. The chloride level was observed to be significantly reduced in some groups, but the lowering in chloride level was small compared to what is seen when aluminium causes mortality (cf Fig. 7 a).

Acclimation

The experiments on acclimation in brown trout are presented in Fig. 7. One group had been acclimated for 34 days at pH 5.0 and at the same total Al regime as in Fig. 6. No hyperventilatory response was seen in trout at this water quality. The reference group had been kept at pH 6.2—6.5. The experiment was started on day 35 when 200 μg/l Al was added, giving a total Al of 360 μg/l.

Five parameters were registered in the experiment: Ventilation frequency, haematocrit and

Table 3. *Ventilation frequency (min^{-1}) and blood parameters measured in Atlantic salmon parr under chronic exposure to aluminium. Average values and standard deviations are given ($n=5$). Haematocrit and chloride are measured on the same fish, ventilation frequency not necessarily so. Significant difference from control is noted by ** for < 0.01 and * for $p < 0.05$ (Wilcoxon rank test). The parr held at pH 5.5 are the control group (day 27). Range of Al-concentrations are given for the last 3 days (μg/l).*

Day	pH	Al	Vent. freq.	Haematocrit	Cl^-mM
27	5.5	110—170	77±11.7	33±3.2	130± 3.6
29	5.5	230—250	121±10.7**	43±8.4*	118±12.5
9	5.3	130—180	98±13.9*	43±4.1*	122± 5.3
11	5.3	140—180	103±14.1*	42±0.7**	126± 7.0
27	5.3	110—170	99±13.5*	43±5.8*	112± 8.2**
29	5.3	230—250	122±10.1**	44±4.0*	112± 6.6**

plasma values for Na, Cl and osmolarity. It is evident from Fig. 7 that, for all the parameters, the reference group shows a more acute response than the acclimated group: Hyperventilation and haematocrit increase is earlier and ion and osmolarity loss rates are faster. At this level of exposure (pH 5.0 and 360 μg/l Al) the lethal level is exceeded even for the acclimated group, and differences between acclimated and control might therefore have been greater at a more moderate stress level.

The acclimation experiment shows that fish living in a "good" water quality are more vulnerable during rapid, shortlasting changes in water quality. It stresses the need to monitor water quality in the field more or less continuously to be able to link disappearance of fish populations to acidification of the water. It also explains why data for toxic Al/pH-combinations, obtained in tank experiments, are of limited value in interpreting field data.

Mode of action

Our fish were observed to cough and hyperventilate, and excessive mucous clogging was registered on the gill surface. This is in agreement with earlier findings (Muniz and Leivestad, 1980 a; Rosseland 1980). The increase in ventilation rate (and coughing frequency) can be a result of an irritant effect of aluminium, causing excessive mucous secretions and impaired gas exchange across the gill. Reduced venous oxygen tensions have been found in fish exposed to Al (Muniz and Leivestad 1980 b).

Catecholamine release can be initiated by hypoxia or chemical irritation of the gill tissue (Hughes 1981). Catecholamine release will tend to increase the recruitment of perfused secondary lamellae and increase blood-flow through the gill, and this may add an osmotic disturbance to the fish. Increased blood perfusion of the lamellae combined with increased ventilation may increase the permeability of the gill to water and salt, inducing water gain and loss of salt from the hyperosmotic fish (Randall 1982). The rapid loss of salt seen in toxicity experiments with aluminium may therefore partly be secondary to changes in ventilation/perfusion, induced by irritation of gill tissue. Since it has been shown that increased calcium levels will reduce the loss of salt in aluminium-stressed fish (Muniz and Leivestad 1980 a) the gill permeability for sodium may be affected directly by Al in a similar manner as in hydrogen ion stress (McWilliams 1980). In recent experiments it has also been shown that the gill Na-K-ATP-ase, responsible for active sodium uptake, is blocked by Al at pH 5 (Kjartansson 1984).

The mode of toxic action of aluminium is certainly very complex, and further studies are needed. This involves both the physiological responses involved, the effects of acclimation and the relative toxicity of different Al-species.

III. REFERENCES

Baker, J. P. and C. L. Schofield. 1980. Aluminum toxicity to fish as related to acid precipitation and Adirondack surface water quality. p. 292—293. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

— and C. L. Schofield. 1982. Aluminum toxicity to fish in acidic waters. Wat. Air Soil Poll. 18: 289—309.

Bersillon, J. L., D. W. Brown, F. Fiessinger and Y. D. Hem. 1978. Studies of hydroxyaluminium complexes in aqueous solution at 25°C. Geochem. Cosmochem. Acta 43: 861—868.

Cronan, C. S. and C. L. Schofield. 1979. Aluminium leaching response to acid precipitation: Effects on the high-elevation watersheds in the Northeast. Science 204: 304—307.

Dickson, W. 1980. Properties of acidified water. p. 75—83. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

Dougan, W. K. and A. L. Wilson. 1974. The absorbtiometric determination of aluminium in water. A comparison of some chromogenic reagents and the development of an improved method. Analyst, Camb. 99: 413—430.

Driscoll, C. T. 1980. Aqueous speciation of aluminum in the Adirondack region of New York State, USA. p. 214—215. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo —Ås.

— J. P. Baker, J. J. Bisogni Jr and C. L. Schofield. 1980. Aluminium speciation and equilibria in dilute acidic surface waters of the Adirondack region of New York State. Presented before the Div. Environm. Chem., Am. Chem. Soc., Las Vegas, Nevada March 28 — April 2. Contr. Upstate Freshw. Inst. 7.

Fivelstad, S. 1983. Aluminium complexes and their toxic effects on salmon (*Salmo salar* L.) and brown trout (*Salmo trutta* L.). Thesis, Univ. Bergen, Norway. 133 p.

Grande, M., I. P. Muniz and S. Andersen. 1978. Relative tolerance of some salmonids to acid waters. Verh. internat. Verein. Limnol. 20: 2076—2084.

Hughes, G. M. 1981. Effects of low oxygen and pollution on the respiratory systems of fish. p. 121—146. *In* Stress and Fish. Ed.: A. D. Pickering. Academic Press, London—New York.

Jensen, K. W. and E. Snekvik. 1972. Low pH wipe out salmon and trout populations in Southernmost Norway. Ambio 1: 223—226.

Kjartansson, H. 1984. The effect of exposure to low pH (5) and aluminium on the specific Na^+ —K^+ —(Mg^{++}) —ATPase and residual Mg^{++} —ATPase activities in tissue homogenates and on plasma chloride concentration in Atlantic salmon, *Salmo salar*, presmolt and smolt, and char, *Salvelinus fontinalis*, yearlings. The effect on plasma cortisol concentration in *Salmo salar* smolt. Thesis. Univ. Bergen, Norway. 92 p.

May, H. M., P. A. Helmke and M. L. Jackson. 1979. Gibbsite solubility and thermodynamic properties of hydroxy — aluminium ions in aqueous solution at 25°C. Geochem. Cosmochem. Acta 43: 861—868.

McWilliams, P. G. 1980. Acclimation to an acid medium in the brown trout (*Salmo trutta*). J. exp. Biol. 88: 269—280.

Muniz, I. P. and H. Leivestad. 1980 a. Acidification effects on freshwater fish. p. 84—92. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

— and H. Leivestad. 1980b. Toxic effects of aluminium on the brown trout, *Salmo trutta* L. p. 320—321. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

Randall, D. J. 1982. Blood flow through gills. *In* Gills. Eds.: D. F. Houlihan, Y. C. Rankin and T. Y. Shuttleworth. Exp. Biol. Sem. Ser. 16: 173—193.

Rosseland, B. O. 1980. Physiological responses to acid water in fish. 2. Effects of acid water on metabolism and gill ventilation in brown trout, *Salmo trutta* L. and brook trout, *Salvelinus fontinalis* Mitchill. p. 348—349. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

Schofield, C. L. and J. R. Trojnar. 1980. Aluminium toxicity to brook trout (*Salvelinus fontinalis*) in acidified waters. p. 341—365. *In* Polluted rain. Eds.: T. Y. Toribara, M. W. Miller and P. E. Morrow. Plenum Press, New York.

Smith, R. W. 1971. Relation among equilibrium and nonequilibrium aqueous species of aluminium hydroxy complexes. Nonequilibrium systems in natural waters chemistry. p. 250—279. *In* Advances in chemistry. Ser. 106. Am. Chem. Soc., Washington, D.C.

Ecosystem Shifts and Reintroduction of Arctic Char (*Salvelinus salvelinus* (L.)) after Liming of a Strongly Acidified Lake in Southwestern Sweden

BENGT HASSELROT, B. INGVAR ANDERSSON and HANS HULTBERG

Swedish Environmental Research Institute, P. O. Box 5207, S-402 24 Gothenburg, Sweden

ABSTRACT

Before undergoing lime treatment in December 1979 Lake Stora Holmevatten was a strongly acidified oligotrophic lake. Much of the biota including all fish species, had disappeared during the acid period. The indigenous Arctic char population which became extinct, had represented a unique genetic stock, classifying this lake with high national interest.

After lime treatment, pH and alkalinity were observed to increase substantially, and the high aluminium levels (500 $\mu g \cdot l^{-1}$) decreased to less than 100 $\mu g \cdot l^{-1}$. Biomass levels of phytoplankton, zooplankton and benthic organisms increased within one to three years, following liming. Diversity of zooplankton and benthic fauna increased but the diversity of phytoplankton did not change significantly. In contrast to earlier studies, chironomid biomass increased following liming, suggesting that aluminium toxicity may have affected the larvae during the acidified period. The generally increased biomasses and shifts in phytoplankton and zooplankton species indicate that a higher net-production was one important effect of liming.

The introduced Arctic char preyed on a variety of food items, one of which, the introduced *Asellus aquaticus*, was of growing importance. The growth rate of the introduced Arctic char was higher than that of the previous, now extinct population. Occurrence of two-summer-old char in test catches in November 1982 showed that natural spawning was successful already by autumn 1980.

I. INTRODUCTION

The deterioration of ecosystems by acid deposition is extensive. Damage to stressed aquatic biota, especially fish populations, have been documented by many authors (HULTBERG and STENSON 1970, Almer *et al.* 1974; GRAHN *et al.* 1974, OVERREIN *et al.* 1980, MONITOR 1981, HARVEY *et al.* 1981; HAINES 1981, MUNIZ 1981, LAST 1982, HULTBERG 1984).

In Sweden, 18,000 lakes and 80,000 km of stream are estimated to have endangered or damaged fish and crayfish populations (MONITOR 1981). The increase in deposition of sulphur has been halted or even slightly reversed. Current acid-loading would, however, still increase the number of lakes in which the fish populations are lost.

Small forest lakes supporting populations of Arctic char (*Salvelinus* sp.) are all low buffer lakes and consequently most vulnerable to acid precipitation. Lake Stora Holmevatten is one such lake that lost its indigenous char population (cf MALM 1877) due to acidification between 1967 and 1971 (DICKSON *et al.* 1975).

This paper presents observations on ecosystem shifts and the reactions of reintroduced Arctic char (*Salvelinus salvelinus* (L.)) following the liming of Lake Stora Holmevatten.

II. MATERIALS AND METHODS

Site description

Lake Stora Holmevatten (57° 99' N, 11° 96' W) is situated 109 m. a.s.l. in an area underlaid by granitic bedrock. The area is part of a forested highland about 10—15 km inland from the Swedish west coast. Topography, vegetation, and hydrology are similar to the River Anråseå area described by ANDERSSON *et al.* (1984). Data describing the lake are given in Table 1. Lake Stora Holmevatten is dammed approximately 1.5 m and remnants of an older barrage indicate that the lake has been utilized as a water reservoir for centuries. The bathymethric map (Fig. 1) disclose two major basins (max. depths 25 and 30 m) and one smaller basin (max. depth 17 m) close to the outlet. Shal-

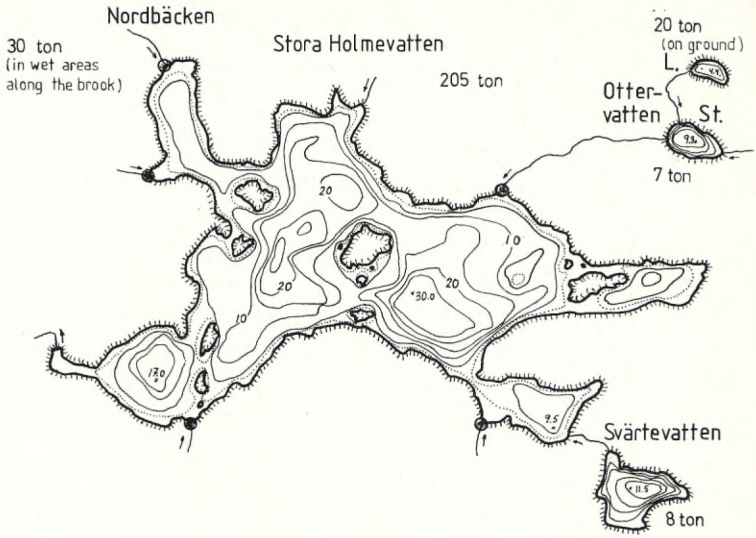


Fig. 1. The watershed of Lake Stora Holmevatten. The depth between isobaths is 2.5 m for the small lakes and 5 m for Lake Stora Holmevatten. In the latter the dotted isobath indicates the depth of 2.5 m. The amount of limestone applied to each area is given in metric tons.

low areas are restricted to the shore-line except for an isolated area in the eastern part.

Liming

During November—December 1979 205 tons of ground limestone (grain size 0—0.5 mm) were spread over Lake Stora Holmevatten. About 50 tons were spread over deep areas and 130 tons along the shore-line. In order to limit acid influx, approximately 4 tons of limestone were blown over the inlet areas of each of 6 small, partly ephemeral brooklets. One brooklet (Nordbäcken) was limed with 30 tons along most of its stretch and 20 tons were spread on the land surrounding Lake Lilla Ottervatten. Including the lime added to Lake Stora Ottervatten and Lake Svärtevatten a total of 270 tons was spread over the watershed of Lake Stora Holmevatten (cf Fig. 1).

Table 1. *Some characteristic data for the Lake Stora Holmevatten system.*

Lake	Watershed area ($m^2 \cdot 10^6$)	Lake area ($m^2 \cdot 10^6$)	$z_{max.}$ (m)	$\bar{z}$ (m)	Lake volume ($m^3 \cdot 10^6$)	Specific runoff ($m^3 \cdot s^{-1} \cdot km^2$)
Lilla Ottervatten	0.035	0.003	4.5	2.3	0.007	0.0165[1]
Stora Ottervatten	0.090	0.006	9.3	4.3	0.026	,,
Svärtevatten	0.080	0.013	11.3	5.2	0.068	,,
Nordbäcken	0.206	—	—	—	—	,,
Stora Holmevatten	1.64	0.284	30.5	9.9	2.8	,,

Lake	Theoretical residence time (year)	Shore line length ($m \cdot 10^3$)	Altitude above mean sea level (m)	Wetlands %	Clear-cut %
Lilla Ottervatten	0.38	0.17	124	∼ 30	0
Stora Ottervatten	0.56	0.28	119	∼ 25	0
Svärtevatten	1.63	0.61	129	2	0
Nordbäcken	—	—	128—109	∼ 25	∼ 5[2]
Stora Holmevatten	3.28	4.66	109	∼ 10	∼ 6

[1] Value is estimated from a corrected annual precipitation of 950 mm.

[2] Most of the area was clearcut during the winter of 1983/84.

Reintroduction of Arctic char

Successful reintroduction of Arctic char was thought to be somewhat difficult when, as in this case, the indigenous population had been lost. In Scandinavia, three sibling species exist (NYMAN *et al.* 1981) and in many northern lakes two or even all three can be found sympatrically (NILSSON 1955, NYMAN *et al.* 1981). The status of the dispersed southern populations have only recently been revealed (NYMAN *et al.* 1981) as exclusively belonging to the species *Salvelinus salvelinus* (L.). The extinct Arctic char of Lake Stora Holmevatten thus was *Salvelinus salvelinus* and the best choice for reintroduction would have been fishes from a similar lake. This was impossible, however, so the Arctic char strain from Lake Vättern was stocked. As both allopatric and sympatric populations show high variation, the introduced fishes were assumed to adapt normally and develop a thriving population, despite their origin.

In June 1980, 79 three-year-old individuals of Arctic char were stocked in Lake Stora Holmevatten. These fish were mature and thus were expected to spawn during their first autumn in the lake. We also introduced 550 yearlings as they might have a better chance for adaption under the prevailing food situation.

Sampling program and physico-chemical analyses

This study began in August 1979. The sampling program was as follows:

Benthic organisms from littoral (3—5 m deep) and profundal (10—15 m deep) bottoms were collected from two stations in May after ice thaw, in August and November. Each time 15 samples were taken randomly by a gravity corer (82 mm diameter) and were sieved through a net (0.6 mm mesh size). Qualitative and quantitative analyses of the plankton community were performed on samples taken four times during the open water season.

Gill-netting was carried out regularly (May, August and November) in order to follow the growth rate of the Arctic char population. Stomach contents of the fishes were sampled at each time.

Physical-chemical analyses of lake and inlet water were carried out 6—8 times a year with a concentration of effort during the ice-free season. At the station with maximum depth, watersamples were taken from a depth of 1 m and 1 m above the bottom. Depth profiles of temperature, oxygen (YSI model 54) and pH (Radiometer pHM 29) were taken. Water samples of 0.5 litre were instantly acid preserved (2.5 ml 5M H_2SO_4) for analyses of Kjeldahl nitrogen and total phosphorus, (Technicon Autoanalyzer). Another 0.5 l sample was brought to the laboratory for measurements of pH, conductivity (Beckman Solumeter), alkalinity (nitrogen purged with MR and BCG as indicators for the titration) and colour (absorption at 400 mm recalculated to mg $Pt \cdot l^{-1}$). Analyses for Ca, Mg, K, Na, Fe, Mn and Al were performed using atomic absorption spectrophotometry (Pye Unicam SP 1900) generally according to the manual of the manufacturer.

Samples from the inlet brooklets were collected and analysed in the same way as for lake water.

III. RESULTS

Water chemistry

The liming in December 1979 increased the pH from about 4.5 to values between 7—8 (Fig. 2). The pH of the surface water varied between 7.0—7.5 during the following four years without any decline. Contrary to this, the deeper water displayed slowly decreasing pH values, probably due to progressively less efficient dissolution of lime deposited in the deeper zones. Liming increased alkalinity and concentrations of calcium, especially in the deeper water (Fig. 3). The reason for this was an unforeseen event. Shortly after lime aplication a heavy storm, which created strong wave action, resuspended lime from the littoral zone. The major part of this resuspended lime was again deposited to the profundal zone. Since lake turnover during 1979 and 1980 was incomplete pH, alkalinity and calcium levels differed widely between the surface and deep waters. Even during the following years, alkalinity and calcium continued to increase in the deep water during the stratification periods. This indicates that limestone dissolution from deeper zones may remain effective over at least four years following the initial treatment.

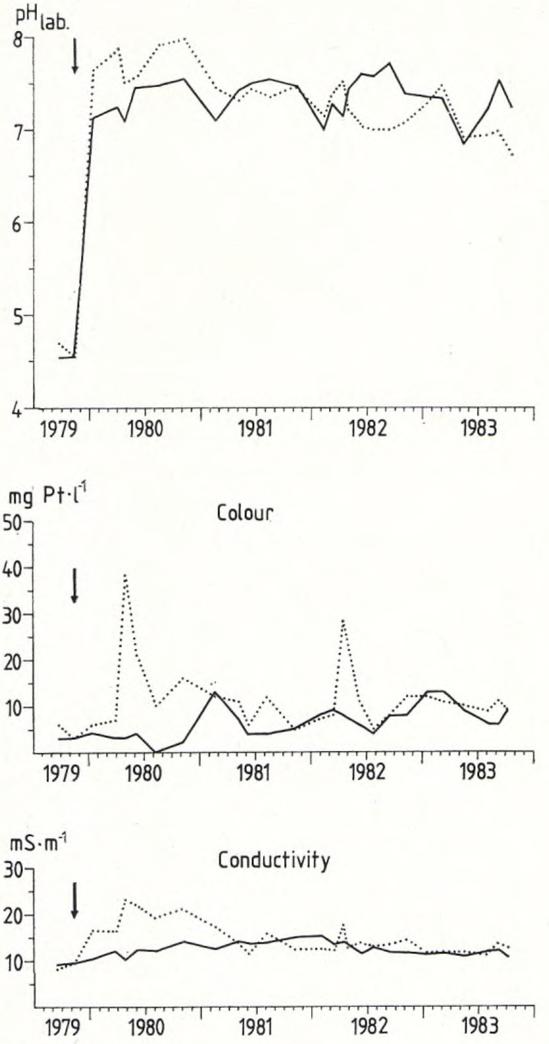


Fig. 2. pH in Lake Stora Holmevatten. Arrows indicate the liming in December 1979. The dotted line is 1 m above the bottom and the smooth line is the surface water (1 m depth).

The manganese concentration in the surface water did not decrease until four months after liming (Fig. 4), suggesting a slow precipitation process. Apart from pH, redox conditions are also important for manganese solubility. The manganese concentrations in deeper water were maximal in late 1980. This was due to precipitation out from surface water coincident with prolonged stratification of deep water. In association with oxygen depletion, a pulse in manganese concentration was observed in the deep water almost every time the water became stratified.

The high levels of total aluminium in association with acid conditions decreased rapidly after liming (Fig. 5). Inflow of aluminium with acid run off in the spring of 1980 counteracted this decrease, but during that summer, aluminium reached a minimum of less than 100 $\mu g \cdot l^{-1}$. During heavy rains in November 1981 and snow thaw in the spring of 1983, the total aluminium concentration increased again both in the epi- and hypolimnion, despite a circumneutral pH.

The average colour value increased in surface water during the four years following liming. This

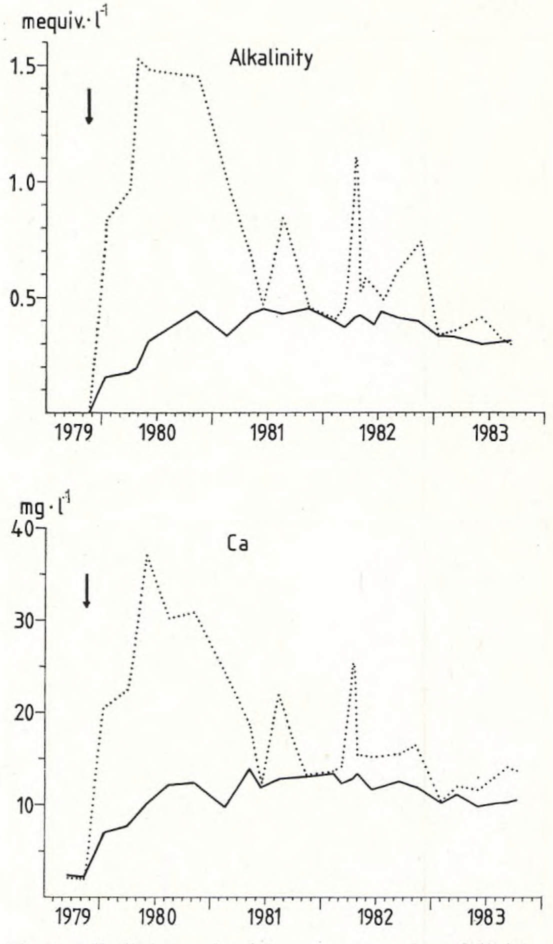


Fig. 3. Alkalinity and calcium concentrations following liming in Lake Stora Holmevatten (same lines as in Fig. 2.).

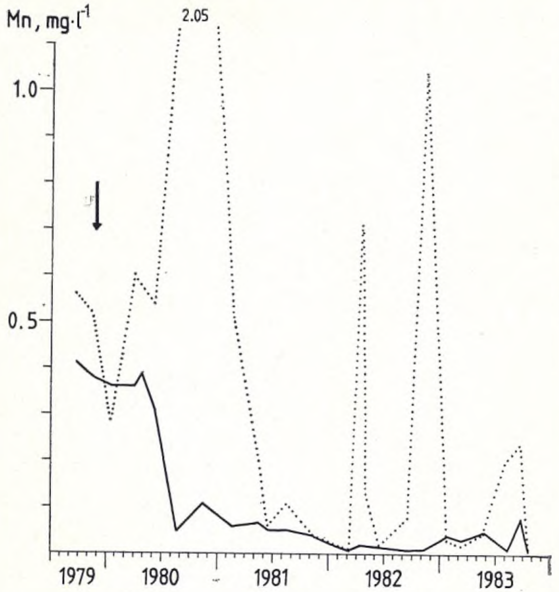


Fig. 4. Manganese concentrations in surface (smooth line) and near-bottom water (dotted line) in Lake Stora Holmevatten.

may be due both to a normalized humus-precipitation instead of the enhanced co-precipitation with aluminium that occurs under acid conditions (Dickson 1980) and a pH-dependent colour increase of fulvic acids. Coincident with this, secchi disc transparency (Fig. 6) decreased from 14 m to 7 m which is comparable to a transparency recorded before acidification in 1948 (Dickson *et al.* 1975). Notable was the sharp decrease in spring 1980, presumably caused by flocks of precipitating aluminium and manganese.

The total phosphorus concentration was very low, both before and after lime treatment (Fig. 7). A minor increase during the summer of 1980 might have been caused by the phosphorus content in dissolved limestone. Furthermore, secondary liming effects like release from sediments and easily degradable organic matter, which had accumulated on the lake bottom during the acid conditions might have contributed.

Total nitrogen concentration increased progressively for about one year following liming, and then became stabilized at a somewhat higher level, than prior to liming (Fig. 7). A notable peak concentration was observed in the deep water in the winter of 1981.

The nitrate-nitrogen concentration in the surface water (Fig. 7) has not changed significantly though. In deep water, a slight increase has occurred. However, during prolonged stratification the concentration was comparatively low.

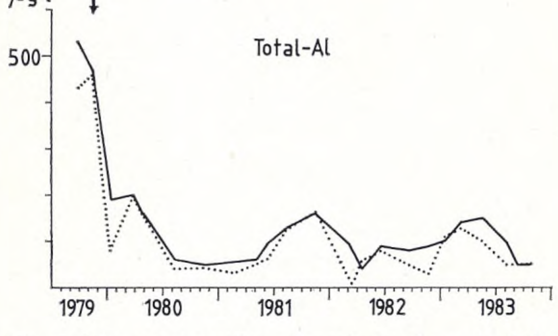


Fig. 5. The concentration of total aluminum in Lake Stora Holmevatten following liming (indicated by arrow).

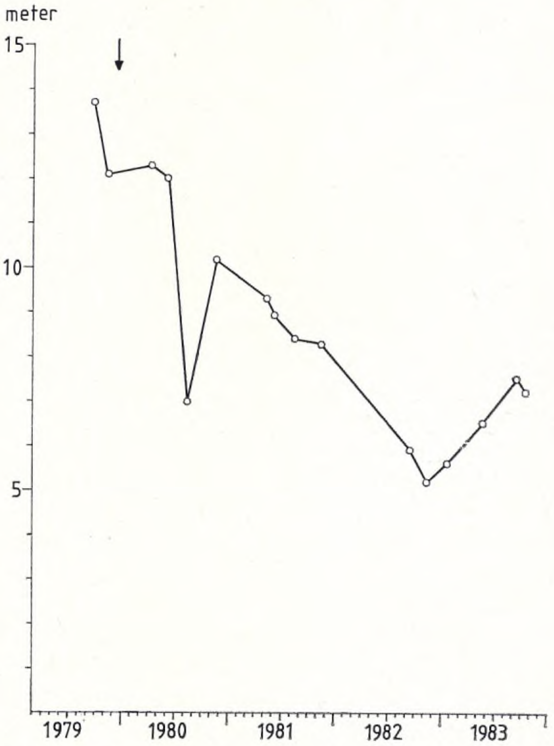


Fig. 6. Secchi disc readings in Lake Stora Holmevatten.

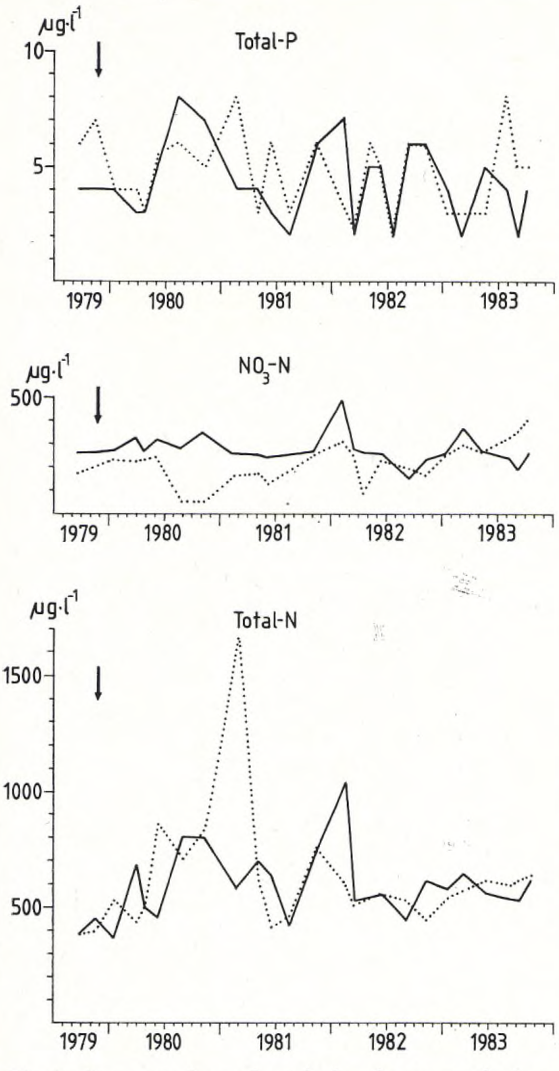


Fig. 7. Concentrations of total phosphorus, total nitrogen and nitrate-nitrogen in surface (smooth line) and nearbottom water (dotted line) in Lake Stora Holmevatten. Arrows indicate the liming.

The deep water oxygen levels in bottom water decreased successively following each lake turnover. Turnover was more or less incomplete between December 1979 and May 1982 (Fig. 8). A partial turnover in the spring of 1981, distributed oxygenated water to the bottom, thus relieving a short period of total oxygen depletion. Effects of the liming on the oxygen conditions are thus unclear, due to lack of pretreatment data. However, the oxygen conditions in the deeper water might have been worsened, since the oxygen depletion in hypolimnic water increased after liming. This was especially marked during the warm summers in 1982 and 1983.

Benthic fauna during acid and limed conditions

The benthic profundal fauna was very sparse during acid conditions. Total biomass was less than 0.2 $g \cdot m^{-2}$. The profundal fauna was dominated by larvae of chironomids (40—70 % of total biomass). *Cyclops* sp. (20—25 %) and a trichopteran belonging to the family Phryganeidae (12 %) were also significant components of the profundal fauna as was *Eurycercus* sp. (5—30 %). Other cladocerans observed were *Bosmina* sp., *Ilyocryptus* sp., and other chydorids. Nematods were also present in the profundal sediments.

The chironomid fauna was represented by the four groups Chironomini, Tanytarsini, Tanypodinae, and Orthocladinae. Table 1 shows that the group Chironomini dominated before liming.

Prior to liming the littoral benthos was dominated by the neuopteran *Sialis lutaria*, which contributed 43—58 % of the total biomass. Chironomid larvae (38—46 %) were also important and all the four chironomid groups were registered. *Eurycercus* sp. was the dominant cladoceran, as in the profundal zone. Other cladocerans found were *Acantholeberis curvirostris*, *Macrothrix* sp., *Alona* sp. and *Bosmina* sp. The trichopteran *Cyrnus* sp. and dipteran larvae of Ceratopgonidae were observed in the littoral but not in the profundal zone.

Following liming, the biomass of the profundal fauna increased to levels approximating those for circumneutral oligotrophic lakes (Fig. 9). During the first two years following liming, biomass continued to increase. In 1982 and 1983 the total biomass varied more but the average level increased. These variations, however, were due to normal ecological seasonal cycling dependent on life strategies of mainly chironomid species. Generally, the biomass of chironomid larvae is lowest during the summer when most species emerge.

The increased profundal biomass was above all due to an increase in chironomid larvae biomass. The group Chironomini dominated during the two

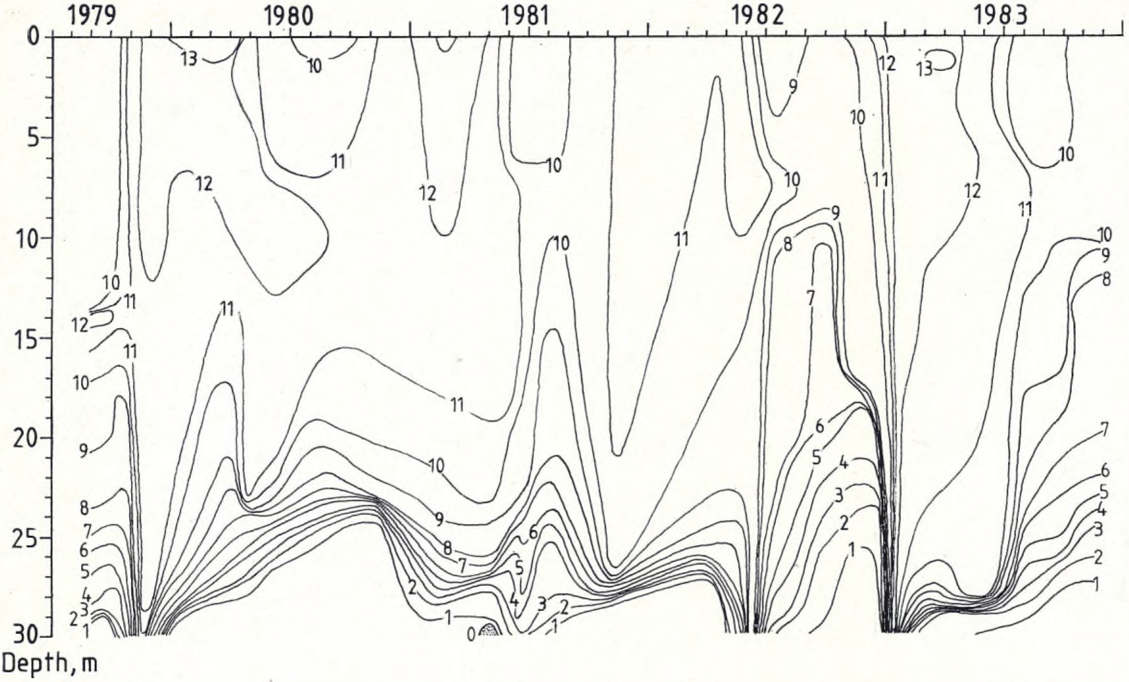


Fig. 8. Isopleths for oxygen concentrations (mg · l^{-1}) in Lake Stora Holmevatten. Liming was performed immediately prior to turnover in November 1979. (Note depletion of oxygen in May 1981).

years following lime treatment. Both number and individual weight increased. In November 1981, the group Tanytarsini increased in abundance and in November 1982 they contributed 86 % of the total number of chironomids (Table 2). At this time, the profundal biomass (8 g · m^{-2}) was the highest recorded. The abundance of the groups Tanypodinae and Orthocladinae generally decreased during the postliming period.

Other profundal fauna elements also changed over several years following the lime treatment. Nematodes disappeared as did *Bosmina coregoni* and *Eurycercus* sp. Others, like *Ilyocryptus* sp., *Cyrnus* sp., and *Corixa* sp. appeared occasionally. Since liming, a few individuals of Ostracoda, Zygoptera and Oligochaeta also have been noted.

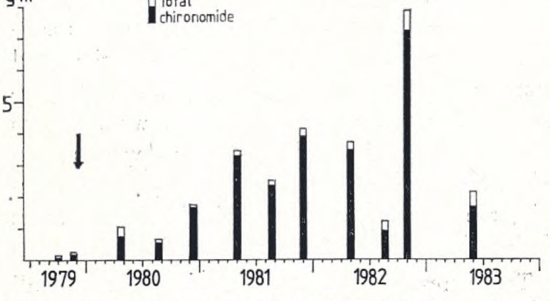


Fig. 9. Biomass (wet weight) of benthic fauna in the profundal sampling area (13 to 16 m). Arrow indicates the liming.

Following liming, the fauna in the littoral zone developed more rapidly than that in the profundal zone. Already in autumn 1980, one year after liming, the littoral biomass had increased substantially (Fig. 10) mainly due to an increase of chironomid larvae biomass. Their relative dominance, however, was significantly less than in the deeper profundal sediments. As was the case in profundal zones, the increased biomass was the result of an increased population of the group Chironomini. However, contrary to the profundal benthos development, Chironomini remained the dominant group throughout the years following liming (Table 2). Only occasionally, the groups Tanypodinae and Orthocladinae increased in abundance. At the end of the study the group Tanytarsini increased, but not as significant as was the case in the profundal zone. Larvae of Ceratopogonidae,

Table 2. *Relative composition and abundance of benthic chironomid larvae in littoral and profundal zone samples through time.*

Date	Chironomini %	Tanypodinae %	Orthocladinae %	Tanytarsini %	Total biomass of chironomids mg · m^{-2}	Percentage chironomid larvae of total biomass
Littoral zone						
Oct. 3, 1979	41	9	36	14	400	46
Nov. 14, 1979	19	22	44	14	1260	38
liming —	—	—	—	—	—	—
April 24, 1980	27	19	50	3	650	54
Aug. 19, 1980	88	4	1	6	2730	69
Dec. 2, 1980	85	9	5	1	8280	63
April 29, 1981	73	5	21	1	5030	75
Aug. 14, 1981	61	36	1	2	510	30
Nov. 26, 1981	76	9	9	6	4110	45
April 26, 1982	63	15	18	4	1620	52
Aug. 18, 1982	46	2	47	5	1730	62
Nov. 3, 1982	81	6	3	10	4460	44
May 19, 1983	83	3	2	13	2520	60
Profundal zone						
Oct. 3, 1979	66	33	—	—	70	40
Nov. 14, 1979	45	16	19	19	160	69
liming —	—	—	—	—	—	—
April 24, 1980	77	6	3	—	720	74
Aug. 19, 1980	77	15	—	8	520	79
Dec. 2, 1980	93	2	4	—	1630	92
April 29, 1981	94	—	10	2	3300	96
Aug. 14, 1981	99	1	—	—	2340	93
Nov. 26, 1981	71	1	4	23	3900	93
April 27, 1982	73	—	12	14	3450	93
Aug. 18, 1982	53	3	6	39	880	72
Nov. 3, 1982	10	—	4	86	7220	93
May 26, 1983	21	—	5	73	1670	79

which also were found prior to liming, increased somewhat following liming.

Following liming the cladocerans *Acantholeberis curvirostris* and *Macrothrix* sp. disappeared, and *Eurycercus* sp. occurred only periodically. In the littoral zone *Bosmina* sp. was found only following the lime treatment. *Alona* sp. was found in small numbers in August 1981.

Asellus aquaticus (about 200 females with eggs) were introduced to the lake in April 1980. Already in August, the same year, *Asellus* occurred in the littoral sampling area (Fig. 10). Sampling during 1981—83 showed that the *Asellus* population spread to other areas and abundance increased. It is not known whether a small number of *Asellus* had survived in the acid lake, although it was not found in spite of intensive search, or whether the introduced individuals seeded the observed colonization.

Zygoptera periodically occurred in relatively high abundance following liming. Ephemerids did not occur in the littoral fauna until November 1981, when the acid tolerant (Johansson and Nyberg 1981) *Leptophlebia vespertina* was observed. By November 1982 its biomass had increased with the coincident appearance of *Centroptilium luteolum*. The biomass of oligochaetes increased slightly following liming and occasionally reached 7% of the total biomass. Nematods occurred both before and after liming at about the same level of abundance.

Individuals of *Corixa* sp. were common in the acidified lake but were generally not found in the bottom zone. In the circum-neutral lake with

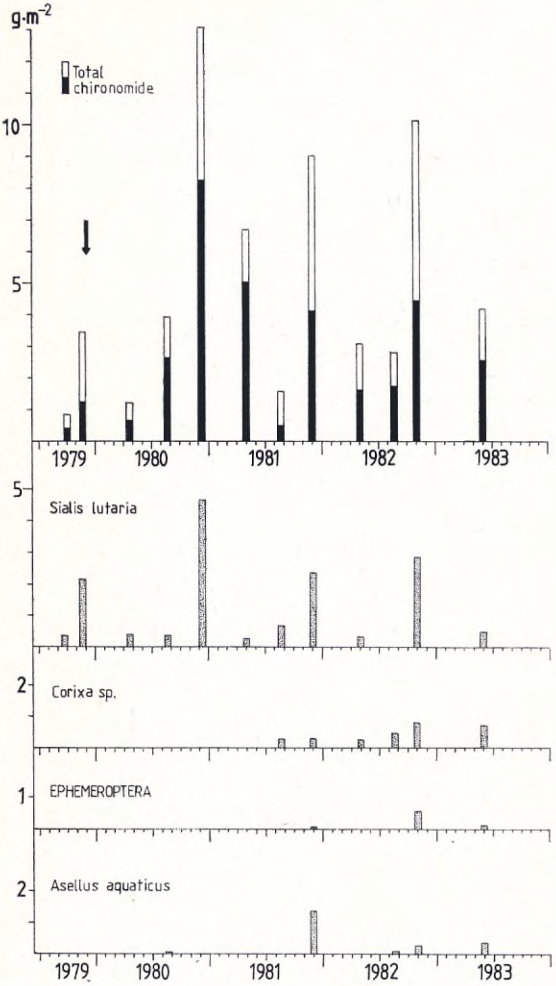


Fig. 10. Biomass (wet weight) of benthic fauna in the littoral sampling area (3 to 7 m). The biomass of dominant non-chironomids is also shown. Arrow indicates the liming.

fish, *Corixa* sp. was found in sediment samples from 1981 (Fig. 10).

Phytoplankton

Twenty phytoplankton species were observed in samples from September 1979. This was a greater number than reported from Lake Stora Holmevatten in 1972, when eleven species were found (Dickson *et al.* 1975). However, in 1979, both the epilimnion and hypolimnion were sampled. The biomass in the epilimnion was about the same in 1972 as 1979 (0.07—0.08 $g \cdot m^{-3}$). In 1972, *Peridinium inconspicuum* and *Gymnodinium uberrimum* were dominant. In September 1979, *Gonyostomum semen* was dominant and contributed 60 % of the total phytoplankton biomass. *Peridinium* sp. also was abundant contributing 36 % of the total biomass. The number of phytoplankton species did not increase following liming. The community underwent a seasonal cycle with 23—25 species during late spring and summer and 11—12 species during winter. The total biomass was observed to increase (Fig. 11) but there was a lack of complete seasonal pretreatment data. The second year following liming did not differ very much from the first year and did not exceed 0.15 $g \cdot m^{-3}$. During the third and fourth years the biomass increased to a higher level (0.3—0.4 $g \cdot m^{-3}$).

The phytoplankton during the first post-liming period (the spring of 1980) was characterized by a sparse flora with *Peridinium* sp. and *Gymnodinium* sp. as dominants. Together they made up

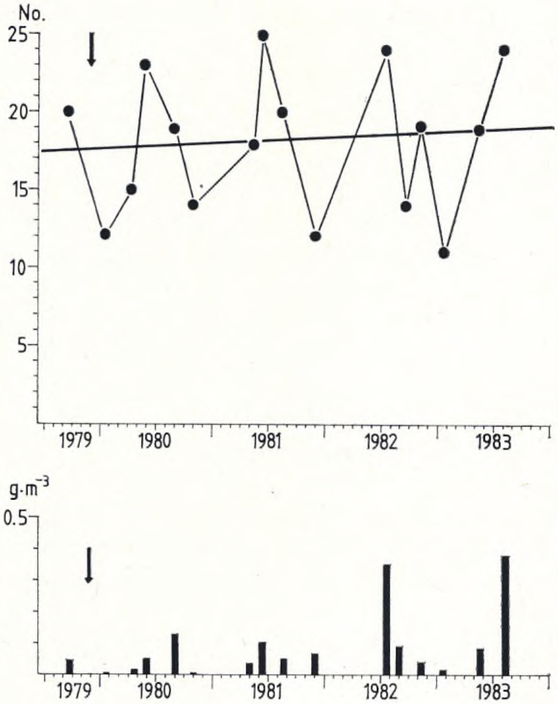


Fig. 11. Number of species and biomass (wet weight) of phytoplankton in Lake Stora Holmevatten. Arrow indicates the liming.

about 90 % of the total biomass. *Gonyostomum semen* which dominated before liming, decreased. In the summer 1980, however, diatoms and also chrysophyceans increased in biomass. The diatoms, *Synedra* sp. and *Tabellaria* represented about 60 % of the total biomass and the chrysophyceans *Bitrichia chodati*, *Chrysolykos* sp., *Dinobryon crenulatum*, and *Stichogloea doederleinii*, contributed another 20 %.

During the second year, some other chrysophyceans, *Dinobryon divergens*, *D. bavarium* and *D. sociale*, together with unspeciated monads increased and became the dominant group (50—60 %). At the same time also, *Cryptomonas* sp. increased to about 40 % of the biomass.

During the third year, the diatom *Cyclotella* sp. reached a biomass of 0.3 g · m^{-3} in July which made up 84 % of the total biomass. For the first time as well, the chlorophycean *Monoraphidium dubowski* increased significantly, although its contribution to the total biomass was small. The chrysophyceans *Chrysidiastrum catenatum* and *Synura* sp. also occurred for the first time in August 1982 at low abundance.

In the autumn of 1982 and in the spring of 1983 the cryptophyceans increased in importance. The species *Croomonas* sp., *Cryptomonas* sp., *Katablepharis* sp. and *Rhodomonas* sp. reached about 40—60 % of the total biomass at this time. During the summer of 1983 the chrysophycean *Dinobryon divergens* again increased notably in abundance and alone reached a biomass of 0.2 g · m^{-3}. Together with *Chrysidiastrum catenatum* and *Ochromonas* sp. they made up the major part of the biomass.

The dinophycean *Ceratium hirundinella* was earlier only observed at low abundance but in August 1983, its biomass increased substantially. Chlorophycean biomass increased in general, first during the third and fourth years. Species like *Monoraphidium dybowski* and *Oocystis* sp., however, occurred only with small biomass levels.

Zooplankton

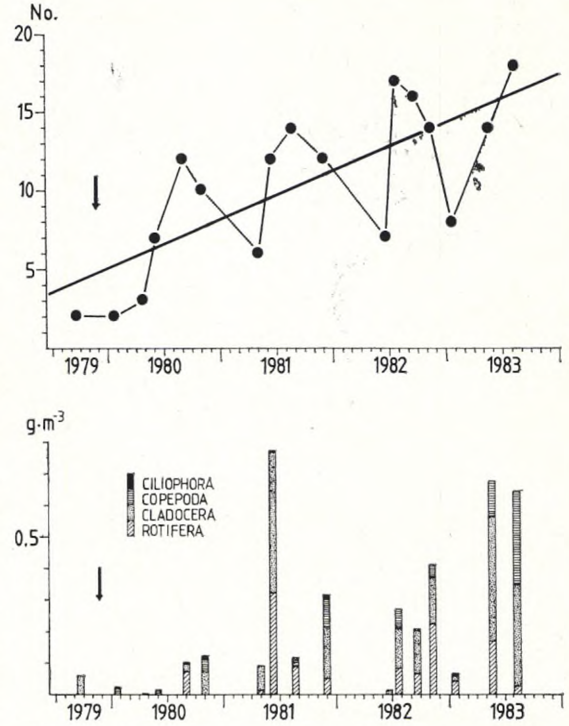


Fig. 12. Number of species and biomass (wet weight) of the zooplankton community in Lake Stora Holmevatten following the liming (arrow).

Only two zooplankton species, *Bosmina longirostris* and *Polyarthra remata* were found in the acidified lake. Their total biomass was very low and was totally dominated by *B. longirostris*. In the earlier investigation in 1972, *Cyclops* sp. and *Bosmina coregoni* were found, the latter being most abundant (Dickson *et al.* 1975).

After liming, a number of zooplankton species successively appeared. The regression line for the number of species was significant ($p < 0.01$) and with a correlation factor $r = 0.74$. Furthermore, a seasonal cycle with few species during winter and spring and a higher number during summer was evident (Fig. 12). During the first year following liming (1980) the biomass increase was insignificant, but during the second year (1981), the summer biomass reached a substantially higher level.

Following liming, the rotifers increased both in number of species and biomass. In August 1980, *Asplanchna priodonta*, *Polyarthra remata* and *Synchaeta* sp. together with *Kellicottia longispina*, *Keratella quadrata* and *Polyarthra vulgaris* represented 76 % of the total zooplankton biomass. *Asplanchna priodonta* seemed to be especially favoured by liming and was the dominating zooplankter during certain periods following liming.

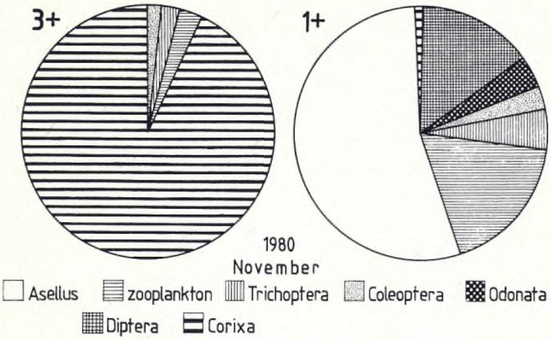


Fig. 13. Comparison of food items found in 1+ and 3+ Arctic char caught in November 1980. Distribution of food item groups is by wet-weight.

In response to the lime treatment cladoceran numbers apparently were enhanced. Both abundance and number of species increased. In August 1980, *Daphnia cristata* was found in hypolimnetic water, but did not occur thereafter. *Bosmina coregoni* and *Bosmina longirostris* dominated the zooplankton in November 1980, and represented 56 % of the biomass. In June 1981 these two species again were dominant, contributing 59 % of the largest zooplankton biomass observed in the lake (0.8 $g \cdot m^{-3}$). Following this, new species such as *Diaphanosoma brachyurum* and *Limnosida frontosa* increased in abundance.

The copepods *Cyclops* sp. and *Eudiaptomus* sp. dominated the sparse zooplankton fauna during the first period following liming. Following this, *Eudiaptomus gracilis* expanded, and in August 1983 it represented about 42 % of the total zooplankton biomass (0.65 $g \cdot m^{-3}$).

Ciliates were not observed in samples until August 1980, well after the lime treatment when *Heliozoa* sp. occurred. Despite increased abundance ciliates never influenced the total biomass significantly. The highest level was reached by *Trachelium ovum* in November 1981, when it represented about 6 % of the total biomass.

The Arctic char population

During the spawning period in November 1980, the first gill netting was performed. In total, thirteen 3+ and twentyone 1+ fishes were caught. The older fish were caught near to shore in shallow (1—4 m) water. Male fish were sexually mature, while females with the exception of one post-spawner, apparently were immature. However, a visual bottom survey revealed that spawning had occurred during autumn. The four-summer-old fish were between 40.5 and 52.9 cm in length and between 610 and 1315 grams in weight.

In the autumn of 1982, only four six-summer-old fish were caught. These measured between 39.8 and 49.0 cm in length, and were 500 and 800 g in weight. They were in very poor condition. Comparing their length and weight at the time of introduction to that at fishing in 1980 and 1982, indicated that these fish had scarcely grown during their period in Lake Stora Holmevatten.

To a very high degree (93 %) the 3+ fish fed on *Corixa* sp. during their first summer in the lake (Fig. 13). The corixids were abundant, because predation in the acidified lake had been virtually absent. The corixid population decreased due to fish predation, and apparently the corixids changed habitats to more sheltered areas. Selecting a narrow food base made these older Arctic char vulnerable to food shortage. Consequently, their

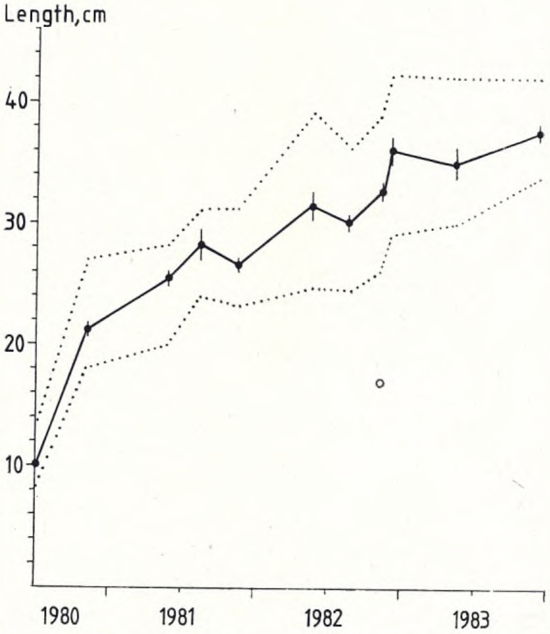


Fig. 14. Mean length of the Arctic char stocked as one-year-old fry in 1980. Bars represent S. E. and the dotted lines give the full range of lengths at each sampling. The circle is the length of two 1+ fish that hatched in the lake.

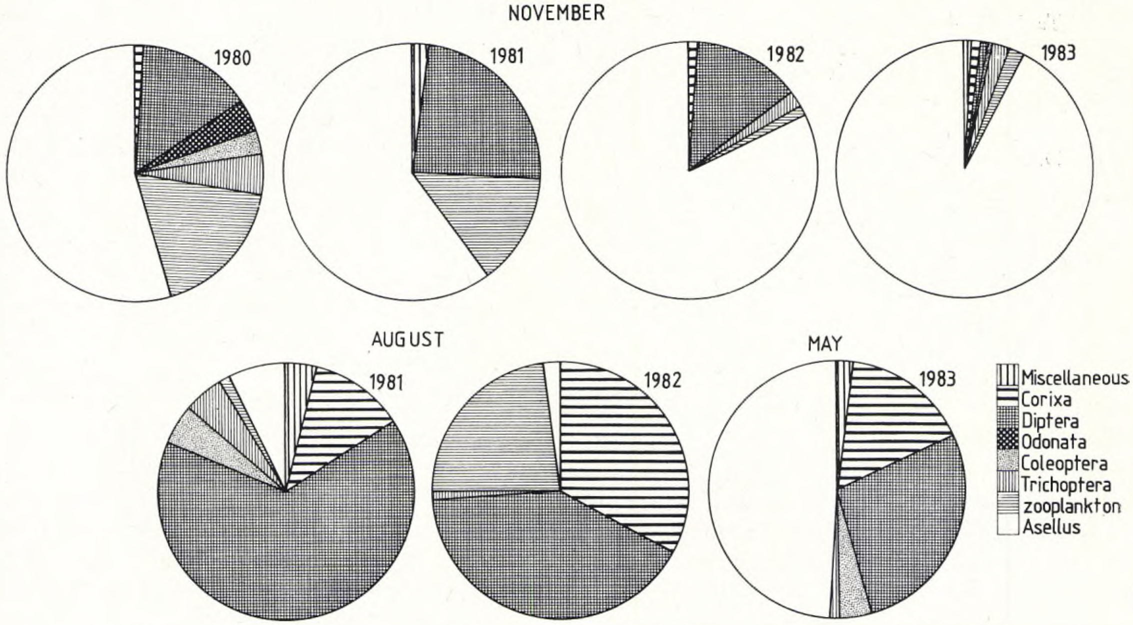


Fig. 15. Food items taken by the same year class (one-year olds in 1980). Distribution of food item groups is by wet-weight.

growth was limited and most of these fish presumably disappeared during 1983.

The one-year-old fish were between 8 and 13 cm when first introduced. By November 1980, their average length had reached about 21 cm. The length increment of this year class from 1980 to 1983 was substantial (Fig. 14). Already by 1982 the fish exceeded the length of the old extinct Arctic char population, which was reported last to be 27—34 cm (Dickson *et al.* 1975). However, by 1983, the growth rate had decreased.

The young fish utilized a more diverse array of food items than the older fish (Fig. 13). However, great differences between individuals were observed. Those fish which preferred *Asellus aquaticus* exhibited a faster growth rate than the others that used other food items. As an example, one fish with its stomach full of *Asellus*, had already by November 1980, reached a length of 27 cm. Other fish, which relied on zooplankton and chironomid larvae, had not grown nearly as much (18—22 cm).

The selection of food items indicated also different habitat preferences between individual fish. The food items varied between different seasons and with increasing age (Fig. 15). Comparing food analyses in November 1980 to that in 1983, shows an increased importance of *Asellus aquaticus*. Despite increased populations and biomass in zooplankton and chironomid larvae, these food items decreased in importance. Fish caught in August, however, generally had a lower percentage of *Asellus* in their stomachs. In the summer fish relied more on dipterans (both larvae and pupae), zooplankton, *Corixa* sp., *Cyrnus* sp., etc.

Food analyses showed that there was an expanding *Asellus* population and an increasing number of fish individuals selecting *Asellus*. The seasonal differences in food choice may have been related to habitat choice which in turn was regulated by water temperature.

IV. DISCUSSION

Previous data suggest that the water chemistry of Lake Stora Holmevatten was typical for oligotrophic forest lakes in this region. By 1948 a pH-value of 6.0 was measured (Dickson *et al.* 1975). By the following decade, acidification had affected the lake, and by 1965 pH was bout 4.5. Coincidently, aluminium and manganese concentrations had increased substantially (0.27 and

Table 3. *The Shannon-Weaver diversity index (H′) for phyto- and zooplankton communities in late summer and early autumn samplings following liming in Lake Stora Holmevatten.*

Date	Phytoplankton community	Zooplankton community
Sep. 24, 1979	1.10	0.345
liming —	—	—
Aug. 29, 1980	1.67	0.841
Aug. 14, 1981	1.08	1.15
Sep. 23, 1982	1.49	1.66
Aug. 9, 1983	1.13	1.94

0.35 $mg \cdot l^{-1}$, respectively). In the autumn of 1979, when we started measurements, pH was still about 4.5. However, aluminium and manganese had increased further to 0.45—0.50 and 0.40—0.55 $mg \cdot l^{-1}$, respectively (cf Figs. 4 and 5).

Lake Stora Holmevatten should thus be regarded as a lake that underwent extreme acidification. Composition and biomass of the aquatic biota also reflected this. Most groups *e.g.* benthos, zooplankton and phytoplankton, had lower biomass compared with other acidified lakes (Hultberg and Andersson 1982, Eriksson *et al.* 1982). Apparently the changes in water chemistry had adversely affected the aquatic biota. Physiological stress or mortality have been experimentally verified in green algae (Hörnström and Ekström 1983), planktonic crustaceans (Potts and Fryer 1979), and chironomid larvae (Havas 1980) at values of pH and/or aluminium that closely correspond to those values measured in Lake Stora Holmevatten.

The aim behind liming was to establish and sustain suitable chemical conditions (circumneutral pH and an alkalinity above 0.1 $meqv \cdot l^{-1}$) for at least a 10-year period. This was assumed to be the necessary minimum time for the lake ekosystem and the reintroduced Arctic char population to achieve a relative stability in both composition and function, without conspicuous treatment effects, thus approaching a natural state (cf Hultberg and Andersson 1982).

Reactions by the aquatic biota to liming were essentially very positive. The biomass of benthic organisms (both on profundal and littoral bottoms), zooplankton, and phytoplankton increased within one to three years.

The diversity of the zooplankton community (calculated as Shannon-Weaver diversity index) increased (Table 3), mostly as a result of an increased number of species. However, the same diversity index for the phytoplankton community did not change significantly following liming.

In contrast to earlier studies (Hultberg and Andersson, 1982, Eriksson *et al.* 1982) the chironomid biomass increased after liming in Lake Stora Holmevatten. Eriksson *et al.* (1982) assumed that the high chironomid biomass often occurring in acid lakes was caused by enhanced food availability that should depend on a more efficient humus precipitation. If so, this effect was counteracted in Stora Holmevatten, presumably by the high levels of both aluminium and manganese. The seasonal variation in chironomid larvae biomass was different from that found by Brundin (1949), with lower values in spring. Reasons for this are unclear but may involve factors like food shortage during winter, predation, and toxic levels of aluminum (as it was most notable in shallow areas).

The number of phytoplankton species was essentially unchanged following the liming, however, species composition changed. Presumably the very low phosphorus content in the lake water both before and after lime-treatment regulated the phytoplankton flora. Similar conclusions have been made by Almer *et al.* (1978) and Eriksson *et al.* (1983). It is difficult, however, to assess the phosphorus turnover associated bioproduction simply by examining phosphorus levels alone. Indication of faster recycling of nutrients (increased total-N in water) and increased bioproduction (increased biomass of zooplankton) is evident in Lake Stora Holmevatten. Furthermore, there has been observed a community shift, from acid-tolerant phytoplankton dominated by dinophyceans to a flora with crypto- and crysophyceans, which have shorter generation times (Eriksson *et al.* 1982). This may indicate that the phytoplankton community turnover is faster thus exhibiting a higher ratio of production to biomass.

Therefore, it seems probable that the observed increased higher phytoplankton biomass following liming, has depended on an increased nutrient supply induced by the liming. This is supported by the fact that some species, which increased only after lime treatment, *e.g. Chrysodiastrum catenatum* and *Dinobryon divergens* have a higher nutrient requirement than acid lake dinophyceans (Hörnström 1979). However, nutrient import from the unlimed catchment has not changed. Hence, nutrients, which were bound to undecomposed organic matter during the acid period, may have been successively recycled and metabolized following liming. Consequently, when this "stored" nutrient supply has been exhausted a stabilization of the bioproduction may follow at a somewhat lower level.

The response of crustaceans and rotifers to the lime treatment was notable. Observed increases were similar to other observations by Hultberg and Andersson (1982), Eriksson *et al.* (1982) and Stenson (pers. comm. 1983). Important food organisms for these groups are planktonic bacteria and phytoplankton such as crypto- and chrysomonads (Pourriot 1977). The abundance of bacterioplankton is not known, but the increase in zooplankton abundance may also be dependent on a detoxification via precipitation of aluminium and manganese to the lake sediments.

Organisms which usually disappear or decrease in abundance following lake liming have often been selected for under the acid conditions due to a change in predation and competition (cf Henriksson *et al.* 1980, Eriksson *et al.* 1980). Examples of such organisms in Lake Stora Holmevatten are the cladocerans *Acantholeberis curvirostris*, *Macrothrix* sp. and the alga *Gonyostomum semen.*

The older (3+) sexually mature Arctic char which were introduced in the lake, fed to a high degree upon *Corixa* sp. The corixids were presumably easy to catch and existed in high abundance as predation was low in the acidified lake. Due to fish predation the pelagic corixid population decreased and coincidently the corixids may have changed habitats, as they were found in both profundal and littoral sediments since the summer 1981. Thus the dependency on pelagic prey by these older Arctic char made them vulnerable to food shortage. Their growth was poor and by 1983 most had probably died. However, the intention behind the stocking of these fishes was successful as two young Arctic char that had hatched in the lake, were caught in November 1982. Otolith analyses disclosed the two fishes to be 1+ year class. Spawning must therefore have been successful in the autumn of 1980.

The Arctic char (1+) stocked in June 1980 selected a more diverse food composition and adapted well to the environment of Lake Stora Holmevatten. They preyed on food items which normally are important for Arctic char, such as larvae and pupae of chironomids, tricopterans and larger zooplankton (Nilsson 1955). However, *Asellus aquaticus* became the most important food during winter. These fish have also reached a length greater than the previous now extinct population. Apparently, they may constitute a good base for a new selfpropagating population.

V. ACKNOWLEDGMENTS

This work was jointly funded by the National Swedish Board of Fisheries and the National Swedish Environmental Research Institute. For carrying out much of the laborious field work and laboratory work we thank Ingemar Alenäs and Inger Torbrink. For zoo- and phytoplankton analysis we thank Roland Bengtsson and Ingrid Aronsson. We are grateful to Olof Filipsson who performed the analyses of Arctic char otoliths. Marie Enghult and Eva Knudsen typed the manuscript. Arno Rosemarin helped correct the English translation.

VI. REFERENCES

Almer, B., W. Dickson, C. Ekström, E. Hörnström and U. Miller. 1974. Effects of acidification on Swedish lakes. Ambio 3: 30—36.

— W. Dickson, C. Ekström and E. Hörnström. 1978. Sulfur pollution and the aquatic ecosystem. p. 271—311. *In* Sulfur in the Environment. Part II. Ecological impacts. Ed.: J. O. Nriagu. John Wiley & Sons. New York.

Andersson, B. I., I. Alenäs and H. Hultberg. 1984. Liming of a small acidified river (River Anråseån) in southwestern Sweden, promoting successful reproduction of sea-trout (*Salmo trutta* L.). Rep. Inst. Freshw. Res., Drottningholm 61: 16—27.

Brundin, L. 1949. Chironomiden und andere Bodentiere der südschwedischen Urgebirgsseen. Rep. Inst. Freshw. Res., Drottningholm 30. 194 p.

Dickson, W. 1980. Properties of acidified waters p. 75—83. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: H. D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

— E. Hörnström, C. Ekström and B. Almer. 1975. Char lakes south of River Dalälven. Inform. Inst. Freshw. Res., Drottningholm (7). 138 p. (In Swedish with English summary.)

Eriksson, F., E. Hörnström, P. Mossberg and P. Nyberg. 1982. Ecological effects of lime treatment of acidified lakes and rivers. Inform. Inst. Freshw. Res., Drottningholm (6). 96 p. (In Swedish with English summary.)

— E. Hörnström, P. Mossberg and P. Nyberg. 1983. Ecological effects of lime treatment of acidified lakes and rivers in Sweden. Hydrobiologia 101: 145—164.

Eriksson, M. O. G., L. Henrikson, B.-I. Nilsson, G. Nyman, H. G. Oscarson, A. E. Stenson and K. Larsson. 1980. Predator-prey relations important for the biotic changes in acidified lakes. Ambio 9: 248—249.

Grahn, O., H. Hultberg and L. Landner. 1974. Oligotrophication — a self-accelerating process in lakes subjected to excessive supply of acid substances. Ambio 3: 93—94.

Haines, T. 1981. Acid precipitation and its consequences for aquatic ecosystem: a review. Trans. Am. Fish. Soc. 110: 669—707.

Harvey, H. H., R. C. Pierce, P. J. Dillon, J. R. Kramer and D. M. Whelpdale. 1981. Acidification in the Canadian aquatic environment. Natural Res. Coun. Canada, NRCL 18427. 369 p.

Havas, M. 1980. A study of the chemistry and biota of acid and alkaline ponds at the Smoking Hills, N. W. T. Ph. D. Thesis. Univ. Toronto, Ontario. 195 p.

Henrikson, L., H. G. Oscarson and J. Stenson. 1980. Does the change of predator system contribute to the biotic development in acidified lakes? p. 316. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

Hultberg, H. 1984. Effects of acid depositions on aquatic ecosystems. p. 167—185. *In* Proceedings of the Symposium of Acid Deposition — a challenge for Europe. Karlsruhe 19—21 Sep. 1983. Eds.: H. Ott and H. Stangl. Comm. Europ. Comm., Dir. Gen. Sci., Res. Development.

— and I. Andersson. 1982. Liming of acidified lakes: Induced long-term changes. Wat., Air Soil Poll. 18: 311—331.

— and J. Stensson. 1970. Effects of acidity on the fishfauna of two small lakes in Bohuslän, southwestern Sweden. Fauna och flora 65: 11—20. (In Swedish with English summary.)

Hörnström, E. 1979. Kalknings- och försurningseffekter på växtplankton i tre västkustsjöar. Nat. Swed. Environm. Prot. Board SNV PM 1220. 45 p. (In Swedish with English summary.)

— and C. Ekström, 1983. pH-, närings- och aluminiumeffekter på plankton i västkustsjöar. Nat. Swed. Environm. Prot. Board SNV PM 1704. 124 p. (In Swedish with English summary.)

Johansson, K. and P. Nyberg. 1981. Acidification of surface waters in Sweden — effects and extent 1980. Inform. Inst. Freshw. Res., Drottningholm (6). 118 p. (In Swedish with English summary.)

Last, F. T. 1982. Effects of atmospheric sulphur compounds on natural and man-made terrestrial and aquatic ecosystems. Agric. a. Environm. (7): 299—387.

Malm, A. W. 1877. *Salvelinus alpinus* (röding). p. 540—544. *In* Göteborgs och Bohusläns fauna. Göteborgs Handelstidnings tryckeri, Göteborg. (In Swedish.)

Monitor. 1981. Försurningen av mark och vatten. Nat. Swed. Environm. Prot. Board SNV Medd. 3. 175 p. (In Swedish.)

Muniz, I. P. 1981. Acidification — effects on aquatic organisms. *In* Beyond the energy crisis opportunity and challenge. Eds.: R. A. Fazzolare and C. B. Smith. Third Int. Conf. Energy Use Manag., Berlin (West), Oct. 26—30, 1981. Vol. IV, A 101—A 123. Pergamon Press, Oxford & New York.

Nilsson, N.-A. 1955. Studies on the feeding habits of trout and char i North Swedish lakes. Rep. Inst. Freshw. Res., Drottningholm 36: 163—225.

Nyman, L., J. Hammar and R. Gydemo. 1981. The systematics and biology of landlocked populations of Arctic char from northern Europe. Rep. Inst. Freshw. Res., Drottningholm 59: 128—141.

Overrein, L. N., H. M. Seip and A. Tollan. 1980. Acid precipitation — effects on forest and fish. Final report of the SNFS project 1972—1980. SNSF Res. Rep. FR 19/80. Oslo—Ås. 175 p.

Potts, W. T. W. and G. Fryer. 1979. The effect of pH and salt content in sodium balance in *Daphnia magna* and *Acantholeberis curvirostris* (Crustacea; Cladocera). J. Comp. Physiol. 129: 289—294.

Pourriot, R. 1977. Food and feeding habits of Rotifera. Arch. Hydrobiol. Beih. Ergebn. Limnol. 8: 243—260.

Lime Influence on Macro-Invertebrate Zooplankton Predators

LENNART HENRIKSON and HANS G. OSCARSON

Department of Zoology, University of Gothenburg, P. O. Box 25059, S-400 31 Gothenburg, Sweden

ABSTRACT

Certain macro-invertebrates believed to be zooplankton predators in the acidified and fishless Lake Gårdsjön were studied before and after a lime treatment. *Chaoborus flavicans* was most abundant and had the highest biomass followed by the corixid species *Glaenocorisa propinqua. Cymatia bonsdorffi* and four different species of dytiscids were less abundant. All groups in the study increased their abundance after the lime treatment, probably as a consequence of improved prey availability. *Chaoborus* increased more than *Glaenocorisa* which enhanced the significance of the *Chaoborus* population after the liming. The open-water predators showed a clear horizontal habitat separation which was not changed after the lime treatment; *Chaoborus flavicans* occurred in the deepest part of the lake farthest from the shore, while *Cymatia bonsdorffi, Chaoborus obscuripes* and the dytiscids mainly occupied the most shallow habitat. *Glaenocorisa* utilized an intermediate habitat. The habitat separation is probably a result of adaption to an environment with fish predators, choice of prey and interactions within the invertebrate predator community.

I. INTRODUCTION

In most lakes, fish is the dominant zooplankton predator. It is a size selective visual dependent predator and its feeding strategy can result in a reduced mean body size of the zooplankton community (DODSON 1974, STENSON 1976). For some species of zooplankton, for example *Holopedium gibberum*, an increased pressure from fish predators can also significantly reduce the abundance (*e.g.* Henrikson *et al.* 1984, in the press).

There are also macro-invertebrate groups that can influence the zooplankton community. For example *Chaoborus* which is a potent zooplankton predator can reduce the abundance of several crustacean species (NEILL 1981). Other taxonomical groups which have been named as potential zooplankton feeders are Dytiscidae (ARTS *et al.* 1981) and Corixidae (NYMAN *et al.* 1984, in the press). In an ordinary Swedish oligotrophic lake, a well established fish population can constrain both abundance and distribution of many species of the invertebrate groups mentioned above (*e.g.* STENSON 1981, HENRIKSON and OSCARSON 1978 a, HENRIKSON and OSCARSON unpubl.). It is, therefore, probable that the influence on the zooplankton structure from insect predators is of minor importance in this kind of lake.

In lakes where the acidification process is advanced, the fish population is heavily reduced with regard to both number of species and abundance, and many lakes become more or less empty of fish. In such lakes, macro-invertebrate predators which were previously missing or in low numbers in unsheltered habitats because of their susceptibility to fish predation, can increase in number.

This is the case with *e.g. Chaoborus obscuripes* which is more sensitive to fish predation than *Chaoborus flavicans*, probably because of different behaviour patterns (STENSON 1981) and with the corixid species *Glaenocorisa propinqua* (HENRIKSON and OSCARSON 1981). The latter, like *Chaoborus*, is a zooplankton feeder which utilizes the pelagic habitat in acidified lakes with weak or missing fish populations (HENRIKSON and OSCARSON 1984, in the press). If such an acidified lake with well-established populations of different macro-invertebrate predators is limed, the remaining fish population, if any, will reproduce. This will lead to an increased predator pressure from fish and thereby to a reduced population of large invertebrate species. Thus, the pelagic predator-prey system will go back to a fish-zooplankton system which is more normal for these kinds of oligotrophic lakes. The changed fish predator influence on the zooplankton community of a limed acid lake has been demonstrated by Henrikson *et al.* 1984 (in the press).

In limed lakes without remaining fish and where no fish are reintroduced after the lime treatment, the pelagic predator-prey system will continue to rely on relations between insects and zooplankton. In this study, we will present the macro-invertebrates that are potential zooplankton predators of the acidified Lake Gårdsjön and discuss their development after a lime treatment.

II. STUDY AREA AND METHODS

The lake that we have studied, Lake Gårdsjön, is situated on the Swedish west coast in an area highly subjected to acid pollutants. In the same catchment area lies Lake Stora Hästevatten which has been used as a control (Fig. 1). Lake Gårdsjön is 31 hectares and has a maximum depth of 18.5 metres and Lake Stora Hästevatten is 3.3 hectares with a maximum depth of 6.5 metres. The lakes have been acid for several years and are devoid of fish. Both lakes are oligotrophic with sparse stands of water vegetation. In April 1982, Lake Gårdsjön was limed ($CaCO_3$) while Lake Stora Hästevatten remained untreated. Characteristic chemical-physical data before and after the lime treatment are given in Table 1.

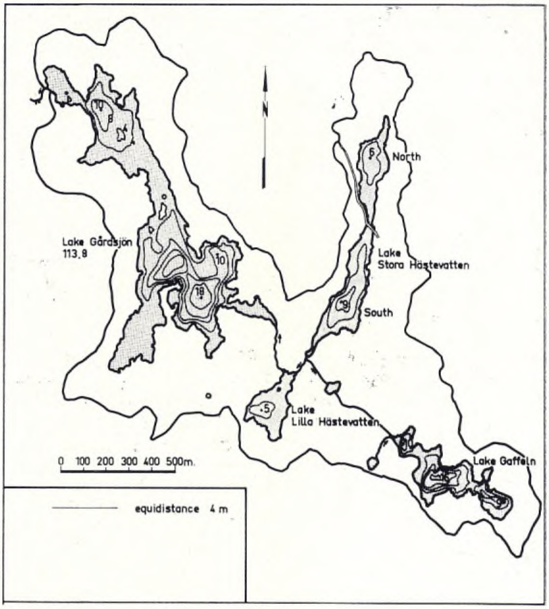


Fig. 1. Study area.

As this study is a part of a project dealing with ecological effects of acidification and liming, further data about Lake Gårdsjön and the surroundings are published elsewhere (Andersson and Olsson 1984).

The sampling started during the autumn 1978 and autumn sampling continued for four years

Table 1. *Some physical and chemical parameters and the chlorophyll a content in Lakes Gårdsjön and Stora Hästevatten. Minimum and maximum epilimnic values from 17—20 sampling occasions per year.*

	Lake Gårdsjön		Lake Stora Hästevatten	
	before liming 1980—81	after liming 1982—83	1980—81	1982—83
pH	4.5—4.8	7.0—7.9	5.0—5.5	4.9—5.6
Alkalinity ($meq \cdot l^{-1}$)	0	0.20—0.54	0—0.01	0
Conductivity ($mS \cdot m^{-1}$)	5.7—7.2	7.5—10.5	4.8—6.2	5.5—6.7
Colour ($mg\ Pt \cdot l^{-1}$)	0—12	1—10	0—10	1—13
Secchi depth (m)	6.5—13.0	2.4—7.9	$>$ 6.5	$>$ 6.5
Total phosphorus ($\mu g \cdot l^{-1}$)	1—12	3—9	2—6	2—5
Nitrate ($\mu g \cdot l^{-1}$)	30—180	23—162	30—120	30—130
Ammonium ($\mu g \cdot l^{-1}$)	6—127	7—260	0—68	0—51
Total nitrogen ($\mu g \cdot l^{-1}$)	221—648	229—540	185—495	90—540
Calcium ($mg \cdot l^{-1}$)	1.6—2.4	7.3—13.7	2.0—3.7	2.2—2.6
Sulfate ($mg \cdot l^{-1}$)	7.8—13.0	8.5—11.2	6.7—11.4	8.8—10.3
Aluminium ($mg \cdot l^{-1}$)	0.10—0.49	0—0.48	0.01—0.45	0.03—0.77
Chlorophyll *a* ($\mu g \cdot l^{-1}$)	0—2.8	0.6—6.2	—	—

before and two years after the lime treatment. The samples were taken during the autumn as the populations of the predators are uniform and relative stable at this time, since the larval development is finished. Populations of the fourth instar of *Chaoborus flavicans* were estimated by bottom sampling with a core sampler with an opening area of 56 cm^2. 35 sampling sites at depths greater than four metres were randomly chosen, fifteen sites in the northern part and twenty sites in the southern part of Lake Gårdsjön. In the control, Lake Stora Hästevatten, twenty sampling stations were chosen according to the same procedure. All samples were sieved through a 0.25 mm mesh and preserved in 4 % formaldehyde. The estimations of abundance and biomass of *Chaoborus flavicans* are based on these samples. Beside the profundal samples, sampling sites for bottom sampling in the littoral zone (0—4 m) were randomly chosen with fifteen samples in the southern part of Lake Gårdsjön, fifteen samples in the northern part and fifteen samples in Lake Stora Hästevatten. Littoral bottom sampling were performed for two years before and two years after the lime treatment. We also looked for *Chaoborus* in the limnetic zooplankton samples. *Chaoborus obscuripes* was sampled from the shore with standardized sweep net hauls. Twenty hauls were taken from each lake at randomly chosen sites. All samples were preserved in 4 % formaldehyde. The littoral bottom samples were also used to get information about *Chaoborus obscuripes*.

Corixids and dytiscids were sampled with a funnel trap (HENRIKSON and OSCARSON 1978 b) with a sampling area of 0.65 m^2. The funnel encloses a certain volume of water, which depends on the actual water depth from the bottom to the surface at the sampling site. When the animals try to reach the surface in order to replenish their air supply they are forced into a plastic cone and trapped. The action of this sampler is limited to a water depth of about ten metres. Therefore, the sampling sites were randomly chosen within the ten metre isobat although our deepest sampling sites actually were situated at eight metres. All samples were preserved in 4 % formaldehyd.

When we statistically tested the values for change in abundance after the lime treatment, we compared the mean value from each sampling station from the four sampling occasions before the lime treatment with corresponding values from the two occasions after the lime treatment. When we calculated the density of the different predator groups at different water-depth intervals we used all autumn samples from a certain depth interval.

Preserved specimens of *Glaenocorisa propinqua* and *Chaoborus flavicans* were weighed after drying between two paper sheets (WINBERG 1971). This weight was regarded as wet-weight and according to WINBERG this wet-weight differs only about two per cent from the wet-weight of fresh specimens. As the animals had been in formaldehyde for varying periods of time, we tested if this could affect our results. Corixids that we had weighed in 1979 and then preserved again, were weighed four years later and no difference was detected ($\bar{x}(1979) = 19.9$ mg, $SE = 0.51$, $\bar{x}(1983) = 19.8$ mg, $p = 0.50$; not significant with t-statistics). For *Glaenocorisa propinqua*, the biomass was calculated from mean wet-weight values from each year. For *Chaoborus flavicans* we divided the animals in only two categories; animals caught during acid conditions and animals caught during limed conditions.

III. RESULTS

Both *Chaoborus flavicans* and *Chaoborus obscuripes* showed higher autumn abundance of the fourth instar (Fig. 2) after the lime treatment than before ($p < 0.05$, Wilcoxon matched-pair signed-ranks test, SIEGEL 1956). The mean number per sample is about six respective seven times higher for the two species after the liming (Table 2). Highest value of abundance for *Chaoborus flavicans* was reached during the autumn 1982. No such estimate of absolute abundance was possible for *Chaoborus obscuripes* because of the use of a semi-quantitative method. There were no corresponding changes of the two *Chaoborus* species in Lake Stora Hästevatten, the control lake. No *Chaoborus* species were caught neither in the littoral bottom samples nor in the zooplankton net hauls.

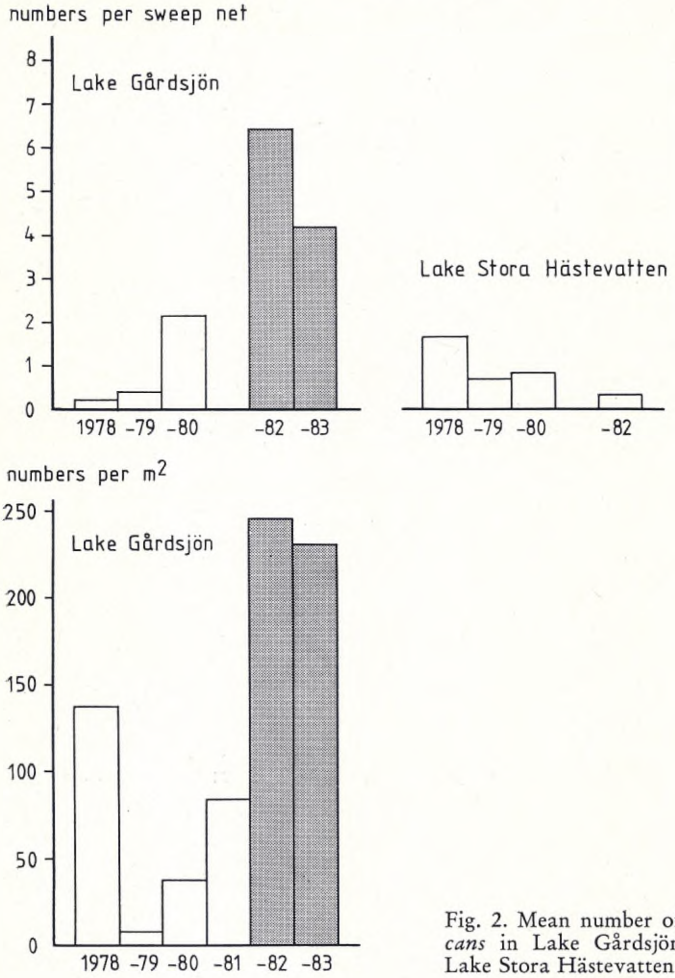


Fig. 2. Mean number of *Chaoborus obscuripes* and *Chaoborus flavicans* in Lake Gårdsjön. Shaded staples indicate limed conditions. Lake Stora Hästevatten serves as a control lake.

The biomass of *Chaoborus flavicans*, (Fig. 3) calculated as wet weight, was also higher after the lime treatment ($p < 0.05$, t-statistics), but the mean weight of single individuals had not changed significantly ($p > 0.05$, t-statistics). During acid conditions *Chaoborus flavicans* was caught only below the ten metre isobat and after the liming below the eight metre isobat. *Chaoborus obscuripes*, however, was only found in sweep net hauls taken around the shore line.

Table 2. *Mean number of animals per sample with standard error (SE) calculated from autumn samples before and after the lime treatment. Wilcoxon matched-pair signed-ranks were used to calculate p-values.*

	Acid conditions			Limed conditions			p	$\frac{\bar{x}_b}{\bar{x}_a}$
	$\bar{x}_a$	SE	n	$\bar{x}_b$	SE	n		
Chaoborus flavicans	0.79	0.27	66	4.57	1.59	46	< 0.05	6.9
Chaoborus obscuripes	0.76	0.32	66	4.54	1.70	27	< 0.05	6.0
Glaenocorisa propinqua	5.38	0.80	64	11.8	4.89	36	< 0.05	2.2
Cymatia bonsdorffi	0.27	0.10	59	1.58	0.51	36	< 0.05	5.9
Dytiscidae	0.27	0.09	41	1.95	0.72	22	< 0.05	7.2

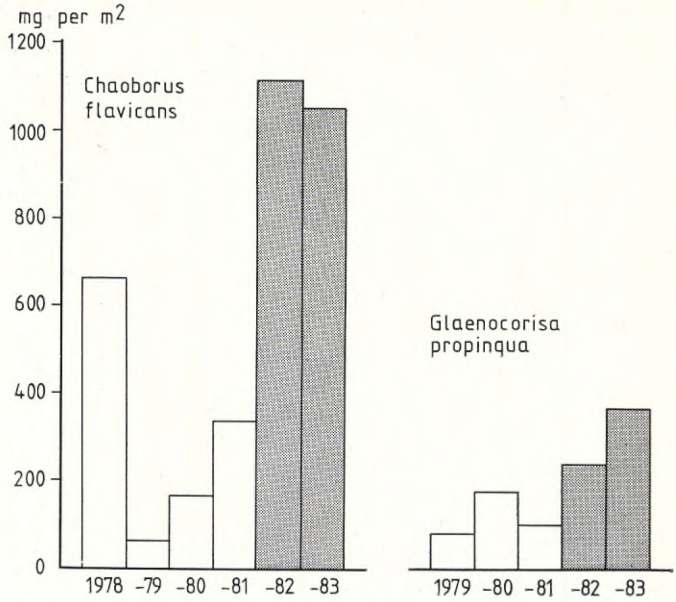


Fig. 3. Autumn biomass (wet-weight) of *Chaoborus flavicans* and *Glaenocorisa propinqua* in Lake Gårdsjön. Shaded staples indicate limed conditions.

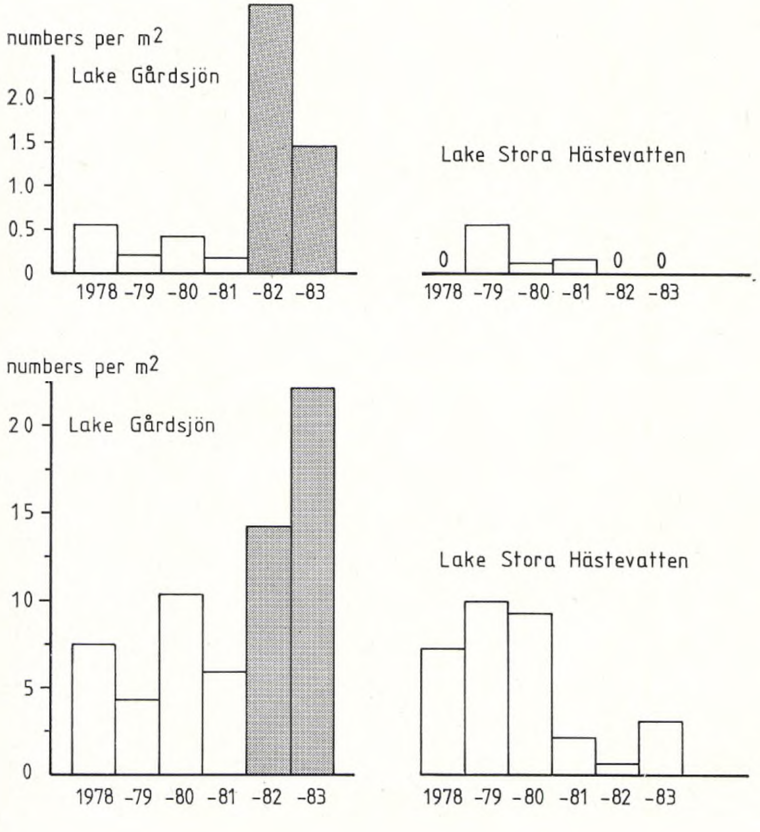


Fig. 4. Mean number of *Cymatia bonsdorffi* and *Glaenocorisa propinqua* in Lake Gårdsjön and Lake Stora Hästevatten. Shaded staples indicate limed conditions.

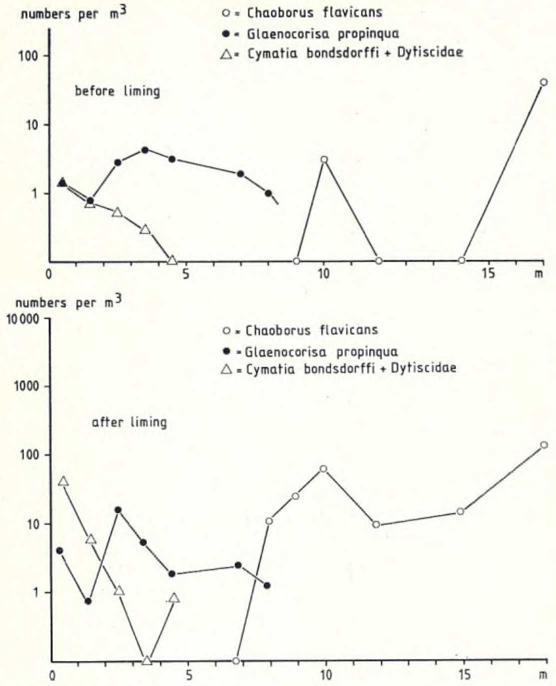


Fig. 5. Density of different predator groups at sampling sites with different water depth. Before and after the lime treatment of Lake Gårdsjön.

The two dominating species of corixids, *Glaenocorisa propinqua* and *Cymatia bonsdorffi* (Fig. 4) were also more abundant after the liming ($p < 0.05$, Wilcoxon matched-pairs signed-ranks test). The mean number per sample was about two respectively six times higher after the liming for the two species (Table 2). During the six years that we sampled corixids the highest abundance was reached in 1983 for *Glaenocorisa propinqua* and in 1982 for *Cymatia bonsdorffi*, *i.e.* after the liming of Lake Gårdsjön. No corresponding changes were noticed in the control lake, Lake Stora Hästevatten. *Glaenocorisa propinqua* was most abundant where the water-depth exceeded two metres (Fig. 5). Because of limitations in the sampling method, we have no quantitive data from sampling sites with a water depth greater than eight metres, but the species was also observed in the middle of the lake where the depth is about eighteen metres. However, *Glaenocorisa* was also found on the bottom samples in water-depth of five to eight metres. Calculated from these samples, the benthic autumn density varied between 0—7.2 individuals per m^2 before the liming and 24.6—31.8 individuals after the liming. *Cymatia bonsdorffi* had its highest density near the shore between zero and one metre depth both before and after the liming.

The wet weight biomass of *Glaenocorisa propinqua* (Fig. 3) increased significantly after the lime treatment ($p < 0.05$, t-statistics). Single individuals of adult males of *Glaenocorisa propinqua* showed no changes in wet weight after the liming ($p \geqslant 0.05$, t-statistics) while there is such a tendency for the females ($0.05 < p < 0.10$, t-statistics). We did not determine individual weights or total biomass for *Cymatia bonsdorffi*.

Besides the two species above, a few specimens of other corixid species as *Arctocorisa germari*, *Sigara distincta*, *Sigara scotti* and *Corixa dentipes* also occurred in the study lake.

Four different species of dytiscids, all smaller than five millimetres were found in samples from Lake Gårdsjön; *Deronectes depressus*, *Laccophilus minutus*, *Coelambus novemlineatus* and *Hygrotus inaequalis*. We have treated the whole group as one. The dytiscids increased significantly in numbers (Fig. 6) after the lime treatment ($p < 0.05$, Wilcoxon matched-pairs signed-ranks test). The population was about seven times greater after the lime treatment (Table 2) with the highest abundance 1983. No corresponding density changes were recorded from the control lake. Like *Cymatia*, the dytiscids had their greatest abundance near the shore at water depth less than one metre and, therefore, we have treated them as one group. No corresponding changes in density occurred in the control lake. We did not determine individual weights or total biomass for the dytiscids.

The horizontal distribution of the different groups in Lake Gårdsjön is depicted in Fig. 5. *Chaoborus flavicans* occupied the deep pelagic part of the lake while *Cymatia bonsdorffi* and the different species of dytiscids were most numerous in the shallow near-shore parts of Lake Gårdsjön. *Glaenocorisa propiqua* seemed to occupy the intermediate area, but because of the lack of samples from deeper parts of the lake we cannot quantify the degree of overlap between *Glaenocorisa propinqua* and *Chaoborus flavicans*. It is not possible to describe the horizontal distribution of *Chaoborus obscuripes* since we do not

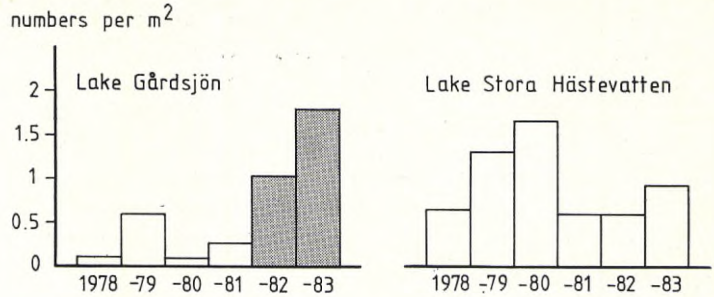


Fig. 6. Mean number of dytiscids in Lake Gårdsjön and Lake Stora Hästevatten. Shaded staples indicate limed conditions.

have suitable samples from open water, but as mentioned earlier, it was found neither in the littoral bottom samples nor in the zooplankton samples.

IV. DISCUSSION

On the basis of our data, we have found that three different taxonomic macro-invertebrates constitute the potential zooplankton predators in the open water community of the acid Lake Gårdsjön; *Chaoborus*, Corixidae and Dytiscidae. All of these free-swimming invertebrate predators increased their abundance after the lime treatment. We suggest that this development is primarily due to two causes; an improved food situation and improved abiotic conditions.

In the case of the first cause, an improved food situation, we know that there was a general increase in phytoplankton biomass, primary production and number of phytoplankton species after the lime treatment of Lake Gårdsjön (LARSSON, unpubl.). The total abundance of zooplankton, did not increase significantly, but there was a shift in species abundance from *Bosmina coregoni* towards *Diaphanosoma brachyurum* (HENRIKSON *et al.* 1984 a, in the press). The measurements of abundance of zooplankton did not indicate any increased prey supply for the insect predators, but with the general improvement of primary production and phytoplankton biomass there is reason to suspect that the zooplankton production also was greater after the lime treatment allowing a higher number of predators. Furthermore, the invertebrate predators, especially those who utilize the more littoral habitats also feed on other prey items than zooplankton. *Cymatia bonsdorffi* is known to feed on active animals such as ephemeropterans for example (WALTON 1943), and in our laboratory studies we found that it can utilize also *Sida crystalina* as prey. In the same laboratory study we found that *Glaenocorisa* was able to feed on *Asellus aquaticus*, ephemeropteran larvae and *Chaoborus* larvae beside different species of zooplankton. Potential prey items such as *Asellus aquaticus* and ephemeropteran and *Chaoborus* larvae, as well as other invertebrate species increased in Lake Gårdsjön after the lime treatment (HENRIKSON *et al.* unpubl.). Thus, we believe that if food were a limiting factor during acid conditions, a general improved availability of different prey items, including zooplankton, might be responsible for the increased populations of invertebrate predators.

Besides the better food environment, the better abiotic environment after liming may have contributed to the increase of free-swimming invertebrates. Invertebrates such as ephemeropterans, plecopterans (RADDUM 1979), *Asellus aquaticus* and molluscs (ØKLAND and KUIPER 1980) as well as different vertebrates such as fish has been found to be sensitive to the chemical-physical conditions they are exposed to in an acid environment. It has been found that even if an animal does not actually die as a result of low pH, its general condition can be affected. This can be manifested in reduced growth rate which has been observed in both fish (MUNIZ and LEIVESTAD 1979) and in larvae of the mayfly *Ephemerella funeralis* (FIANCE 1978). RADDUM (1979) found that different species of stonefly larvae (*Plecoptera*) and caddisfly larvae (*Trichoptera*) have a lower energy content when they were living in acid water than when in non-acid water. In our study, we tried to trace such an impact on the condition of the individuals by measuring the wet-weight of the fourth instar of *Chaoborus flavicans* and adult *Glaenocorisa propinqua* before and after the lime treatment. We found a tendency to (although not statistically significant) an increased mean weight only in *Glaenocorisa propinqua* females. However, corixids are known to be resistent to pH stress (VANGENECHTEN *et al.* 1979) and to other extreme chemical-physical environments such as saltlakes (SCUDDER 1976). Furthermore, *Glaenocorisa propinqua* has been found in a naturally acidified lake where the pH-value was as low as 3.2 (RYDGÅRD 1981). With the knowledge of the tolerance of low pH in *Glaenocorisa propinqua* and our own results from the weighing of *Chaoborus flavicans*, we conclude that it is not pH stress that is acting on the stages we have been working with, at least not of the kind that affects the weight of the animals. Instead we think that the slight increase in body weight which was noticed for *Glaenocorisa propinqua* female can be ascribed to a better food supply.

However, we cannot separate the effects of improved food supply or the improved chemical-physical conditions for the earlier development stages of the different predator species. For example, survival of the first larvae instar of *Chaoborus* has been shown to depend on rotifers. In experiments where food resources for zooplankton were improved by fertilization, it was possible to increase the rotifer survival and thereby improve the survival of the first instar of *Chaoborus* (NEILL and PEACOCK 1980). In Lake Gårdsjön both abundance and number of rotifer species was enhanced after the liming (HENRIKSON *et al.* unpubl.) and the same development has also been described in other limed lakes (HENRIKSON *et al.* 1984 b, in the press.).

It is obvious that there is a horizontal separation of habitats with more or less clear limits between the free-swimming insects in Lake Gårdsjön (Fig. 5). A more detailed study of habitat choice within a species ensemble of corixids in acidified lakes was done by HENRIKSON and OSCARSON 1984 (in the press). Except for a general increase in population density for all the groups we were working with, there were no principal changes in the habitat choice after the lime treatment. There was, however, a tendency for *Cymatia bonsdorffi* and the dytiscids to extend their range to deeper sites farther from the shore, and for *Chaoborus flavicans* to occur at more shallow sites closer to the shore, but we think that this change can as well be explained by a generally increased chance to be caught because of the increased population density. *Chaoborus flavicans* is known to have its daytime habitat in the bottom debris at the deepest part of the lake, probably an adaption to avoid predation from fish as this part of the lake is relatively safe during daytime when most fish predators are hunting. As we can see, the species retains about the same habitat choice even when fish predators are missing, a condition where one would expect a shallower habitat and thus shorter vertical migration distance to be more favourable. But it is not unlikely that this "anti-fish" behaviour may also be adaptive in a fishless environment since *Glaenocorisa propinqua* is a potential *Chaoborus* predator. *Glaenocorsia propinqua* and *Chaoborus flavicans* are temporally separated in the water mass as *Glaenocorisa* is a typical cruising predator searching for prey during daytime (WALTON 1943, HENRIKSON and OSCARSON 1984, in the press) when *Chaoborus flavicans* stays on the bottom in the deepest part of the lake. But according to our results, *Glaenocorisa* also utilizes the bottom habitat down to about ten metres depth which makes it possible that the lack of *Chaoborus flavicans* in shallower water can be explained by predatory influence from *Glaenocorisa.* Since *Glaenocorisa* depends on atmosphaeric oxygen and therefore must visit the water surface to replenish the air store, the lack of the species in samples from greater depths may be explained by a too high cost/benefit relation because to move from surface to great depths must consume a great deal of energy.

Cymatia bonsdorffi and the different dytiscid species dominated the shallow habitat together with *Chaoborus obscuripes.* In laboratory experiments, *Cymatia bonsdorffi* avoided "open water" and instead chose to anchor among weeds (HENRIKSON and OSCARSON 1984, in the press), a behaviour that make them less susceptible to fish predators with which they normally co-exist. According to the same study, there was no predatory interaction between adults of *Glaenocorisa* and *Cymatia* and therefore we ascribe the strong connection to shallow habitat within *Cymatia* more as a result of its food choice and hunting strategy than to predatory impact from *Glaenocorisa propinqua.* The ability for *Glaenocorisa* to prey on *Chaoborus* makes it more plausible that the population of *Chaoborus obscuripes* in the shore-near zone could be influenced by *Glaenocorisa* predation, especially if they should try to expand their habitat to deeper more unsheltered parts of the lake. In our study, however, we had no data supporting this hypothesis.

With exception for *Laccophilus*, the dytiscids in our study has been classified as "poor swimmers", hiding on the bottom or among vegetation (GALEWSKI 1971), which explains their choise of a shallow habitat.

Regarding the different insect groups which constitute the major zooplankton predators, it is probable that their effects on the zooplankton community were of differing importance. *Chaoborus* is a potent zooplankton predator whose influence on the zooplankton structure is well documented. In enclosure studies, NEILL (1981) showed that *Chaoborus* predation affected the abundance of several zooplankton species, for example *Bosmina* which is one of the most common zooplankton species in Lake Gårdsjön (HENRIKSON *et al.* 1984 a) and other acidified lakes. In other enclosure studies (HENRIKSON *et al.* 1984 a, NYMAN *et al.* 1984, in the press, NYMAN *et al.* unpubl.), *Chaborus* predation affected not only the abundance of different zooplankton species, but also reduced the mean body size of both *Bosmina* and *Diaptomus.* FEDORENKO (1975) has estimated the mean percentage of standing crop eaten per day by *Chaoborus* to be two per cent for copepod nauplii, twelve per cent for *Diaptomus* spp and four per cent for *Diaphanosoma.* Similar results were put forward by KAJAK and RANKE-RYBICKA (1970) who reported that seven and thirteen per cent of the zooplankton population were removed daily by *Chaoborus* from two Polish lakes. DODSON (1972) found that *Chaoborus* predation was responsible for 94 per cent of *Daphnia* mortality, which is particularly interesting as *Daphnia* species are missing in Lake Gårdsjön (HENRIKSON *et al.* 1984 a) and other acidified lakes. None were found even after the liming which might indicate that factors other than water quality can be of importance for the occurrence of *Daphnia.* In a recent study, NYBERG (1984) claimed that it is possible that *Chaoborus* can eradicate crustacean species, especially cladocerans, in acid and limed lakes devoid of fish.

Thus, there are many studies that indicate that *Chaoborus* is of vital importance for the zooplankton community, but the interaction between other insect predators and the zooplankton are compared to our knowledge of *Chaoborus* relatively unknown. However, in our enclosure experiments under field conditions, we found that *Glaenocorisa propinqua* was also able to reduce the abundance of prey items such as *Bosmina* and *Eurycercus* (OSCARSON in prep.). Similar results were put forward by NYMAN *et al.* 1984 (in the press). *Cymatia bonsdorffi* has also been reported to utilize zooplankton as prey even if other prey organisms probably are more important (WALTON 1943). In the laboratory, we fed *Cymatia* with other cladocerans as *Sida crystallina.* As for the small dytiscids that were included in our study, we have found very little evidence for their choise of prey. LINDBERG (1944) has in laboratory experiments fed adults of *Hygrotis* and *Deronectes* with benthic organisms as chironomid larvae and with *Daphnia*, but with knowledge of their bad swimming ability and their relatively low abundance, we conclude that their predatory influence are of minor importance for the zooplankton community. However, ARTS *et al.* (1981) have found that larvae of another dytiscid species, *Acilius semisulcatus*, could influence the vertical distribution of a *Daphnia* population. The genus *Acilius* is represented in Lake Gårdsjön (HENRIKSON *et al.* unpubl.) although it was not found in our samples.

Even though we cannot produce absolute estimates of the population density of *Chaoborus obscuripes*, it is probable that together the two *Chaoborus* species were the most important zooplankton predators of the free-swimming insects studied in Lake Gårdsjön. They were the most abundant predator group, even when dytiscids and corixids were treated as one group, and they had also the highest biomass. These relations were even more accentuated after the lime treatment. This conclusion does not exclude the possibility that other predators might be of importance for certain zooplankton species or for local populations.

To summarize our conclusions, we found that in acidified fishless lakes the insect zooplankton predators show a habitat separation which can be a result of adaptations to an environment with fish predators, choice of prey and probably also interactions within the insect predators themselves. The fundamentals of this habitat separation is not affected by the lime treatment, but there is a general increase in predator abundance which we ascribe primarily to an improved prey supply. We also suggest that the structure of the zooplankton community can in large part be a result of the influence of the free-swimming insect predators.

V. ACKNOWLEDGMENTS

This study was carried out within the integrated Lake Gårdsjön project. This work was financially supported by grants from the National Swedish Environmental Protection Board and the Swedish Natural Science Research Council.

VI. REFERENCES

ANDERSSON, F. and B. OLSSON (Eds.) 1984. Lake Gårdsjön — an acid forest lake and its catchment. Ecol. Bull. (Stockholm) 37. (In the press.)

ARTS, M. T., E. J. MALY and M. PASITSCHNIAK. 1981. The influence of *Acilius* (Dytiscidae) predation on *Daphnia* in a small pond. Limnol. Oceanogr. 26: 1172—1175.

DODSON, S. I. 1972. Mortality in a population of *Daphnia rosea*. Ecology 53: 1011—1023.

— 1974. Zooplankton competition and predation: an experimental test of the size-efficiency hypothesis. Ecology 55: 605—613.

FEDORENKO, A. Y. 1975. Feeding characteristics and predation impact of *Chaoborus* (Diptera, Chaoboridae) larvae in a small lake. Limnol. Oceanogr. 20: 250—258.

FIANCE, S. B. 1978. Effects of pH on the biology and distribution of *Ephemerella funeralis* (Ephemeroptera). Oikos 31: 332—339.

GALEWSKI, K. 1971. A study on morphobiotic adaptions of European species of the Dytiscidae (Coleoptera). Polsk. Pismo Entomol. 3: 487—702.

HENRIKSON, L. and H. G. OSCARSON. 1978 a. Fish predation limiting the abundance and distribution of *Glaenocorisa p. propiqua* (Hemiptera). Oikos 31: 102—105.

— and H. G. OSCARSON. 1978 b. A quantitative sampler for air-breathing aquatic insects. Freshw. Biol. 8: 73—77.

— and H. G. OSCARSON. 1981. Corixids (Hemiptera-Heteroptera) the new top predators in acidified lakes. Verh. internat. Verein. Limnol. 21: 1616—1620.

— and H. G. OSCARSON. 1984. Waterburgs (Corixidae, Hemiptera-Heteroptera) in acidified lakes: characteristics, habitat selection and adaptions. Ecol. Bull. Stockholm) 37. (In the press.)

— H. G. OSCARSON and J. A. E. STENSON. 1984 a. Development of the crustacean zooplankton community after lime treatment of the fishless Lake Gårdsjön, Sweden. Rep. Inst. Freshw. Res., Drottningholm 61: 104—114.

— H. G. NYMAN, H. G. OSCARSON and J. A. E. STENSON. 1984 b. Changes in the zooplankton community after lime treatment of an acidified lake. Verh. internat. Verein. Limnol. 22. (In the press.)

KAJAK, Z. and B. RANKE-RYBICKA. 1970. Feeding and production efficiency of *Chaoborus flavicans* MEIGEN (Diptera, Culcidae) larvae in eutrophic and dystrophic lake. Polsk. Arch. Hydrobiol. 17: 225—232.

LINDBERG, H. 1944. Ökologisch-geografische Untersuchungen zur Insektenfauna der Felsentümpel an den Küsten Finnlands. Acta Zool. Fenn. 41: 1—178. (In German.)

MUNIZ, I. P. and H. Leivestad. 1979. Langtidseksponering av fisk til surt vann. Førsøk med bekkerøye *Salvelinus fontinalis* MITCHILL. (Prolonged exposure to fish to acidified water.) SNFS project IR 44/79. Oslo—Ås. 32 p. (In Norwegian with English summary.)

NEILL, W. E. 1981. Impact of *Chaoborus* predation upon the structure and dynamics of a crustacean zooplankton community. Oecologia 48: 164—177.

— and A. PEACOCK. 1980. Breaking the bottleneck: Interactions of invertebrate predators and nutrients in oligotrophic lakes. p. 715—724. *In* Evolution and ecology of zooplankton communities. Ed.: W. C. Keerfoot. Special symposium 3. Am. Soc. Limnol. Oceanogr. University Press of New England, Hanover, New Hampshire and London, England.

NYBERG, P. 1984. Impact of *Chaoborus* predation on planktonic crustacean community in some acidified and limed forest lakes. Rep. Inst. Freshw. Res., Drottningholm 61: 154—166.

NYMAN, H. G., H. G. OSCARSON and J. A. E. STENSON. 1984. Impact of invertebrate predators on the zooplankton composition in acid forest lakes. Ecol. Bull. (Stockholm) 37. (In the press.)

RADDUM, G. G. 1979. Effects on low pH on insect larvae. SNFS project IR 45/79. Oslo—Ås. 58 p. (In Norwegian with English summary.)

RYDGÅRD, M. 1981. Bottenfaunan i den extremt sura Blåmisusjön i Norrbottens kustland. Rep. Dep. Ecol. Zool. Univ. Umeå, Sweden. (Mimeographed in Swedish.)

SCUDDER, G. G. E. 1976. Water-boatmen of saline waters (Hemiptera: Corixidae). p. 263—289. *In* Marine insects. Ed.: L. Cheng. North-Holland Publishing Company.

SIEGEL, S. 1956. Nonparametric statistics for the behavioral science. McGraw-Hill Kogakusha Ltd, Tokyo. p. 75—83.

STENSON, J. A. E. 1976. Significance of predator influence on composition of *Bosmina* spp. populations. Limnol. Oceanogr. 21: 814—822.

— 1981. The role of predation in the evolution of morphology, behaviour and life history of two species of *Chaoborus*. Oikos 37: 323—327.

VANGENECHTEN, J. H. D., S. VAN PUYMBROECK and O. L. J. VANDERBORGHT. 1979. Effects of pH on the uptake of sodium in the waterbugs *Corixa dentipes* (THOMS.) and *Corixa punctata* (ILLIG.) (Hemiptera, Heteroptera). Comp. Biochem. Physiol. 64: 509—521.

WALTON, G. A. 1943. The water bugs (Rhynchota-Hemiptera) of North Somerset. Trans. Soc. Brit. Entom. 8: 231—290.

WINBERG, G. G. 1971. Methods for the estimation of production of aquatic animals. Academic Press, London and New York. 175 p.

ØKLAND, K. A. and J. G. J. KUIPER. 1980. Småmuslinger (Sphaeriidae) i ferskvann i Norge — utbredelse, økologi og relation til forsurning. SNFS project IR 61/80. Oslo—Ås. 85 p. (In Norwegian.)

Development of the Crustacean Zooplankton Community after Lime Treatment of the Fishless Lake Gårdsjön, Sweden

LENNART HENRIKSON, HANS G. OSCARSON and JAN A. E. STENSON

Department of Zoology, University of Gothenburg, P. O. Box 25059, S-400 31 Gothenburg, Sweden

ABSTRACT

The crustacean zooplankton were studied in Lake Gårdsjön (and a control lake) two years before and two years after lime treatment. The lakes were devoid of fish throughout the study. No negative immediate shock effects were noted. No establishment of "new" species occurred, probably as a consequence of invertebrate predation, which was not structurally changed. Despite an increase in phytoplankton biomass and production no increases in total crustacean abundance was noted. Important zooplankton predators, like *Chaoborus* spp and *Glaenocorisa propinqua*, increased significantly and probably reduced the population of *Bosmina coregoni*, which in turn made it possible for *Diaphanosoma brachyurum* to increase its population size. The annual variation in the mean length of *Bosmina* was different from that in lakes with fish populations and it was not changed after liming. On the whole the structure of the crustacean community was similar to that before liming.

The development after liming of a lake devoid of fish will be different from that of a lake with a sparse non-reproducing fish population before liming. In the latter lake the renewed success of fish reproduction will give a large fish fry population which will increase the predation on zooplankton as fish fry are potent zooplankton predators. Hence, changes in the crustacean zooplankton after liming are dependent on the acidification phase, *i.e.* the community structure of both prey and predators before liming.

I. INTRODUCTION

The acidification of freshwater ecosystem is a process where the changed abiotic environment will lead to elimination of certain species. In this new situation, biological mechanisms will in turn lead to further changes in the faunal structure. Predation is such a mechanism which has been shown to be very important for the structure of zooplankton and other communities in lakes (*e.g.* ZARET 1980). Based on the predation situation the acidification of lakes may be divided into different phases, an approach which has been used by HENRIKSON *et al.* (in the press).

Before acidification, predation by fish is dominating and a similar situation can be seen in the early acidification phase when only just extremely sensitive organisms have been eliminated. This situation we call phase 1. When more acid and other components are supplied to the lake, fish and other organisms are gradually affected. Then predatory insects like phantom midge larvae *Chaoborus* spp and waterbug species like the corixid *Glaenocorisa propinqua* (FIEB.) can increase as they are no longer subjected to heavy fish predation (HENRIKSON and OSCARSON 1978, STENSON 1978). Hence, in this phase 2, the predation on zooplankton originates mainly from sparse populations of remaining adult fishes and macroinvertebrates. When all fish have been killed, a great increase in the predatory macroinvertebrates occurs (*op. cit.*) and these exert predatory pressure on zooplankton. This is phase 3. The intensity of predation on zooplankton is probably highest in phase 1 and 3, where there are well established predator populations. Phase 2, an intermediate phase, probably means a low predation intensity. The division of the acidification process into phases may also be based on water quality and faunal changes in different pH-intervals (JOHANSSON and NYBERG 1981, RADDUM and FJELLHEIM 1984, in the press), but we think that for the understanding of the biological development during acidification a predation approach is more useful.

During the last years large-scale lime treatments of lakes have been performed in Sweden in order to mitigate the effects of acidification. The biological response to a lime treatment is to a great

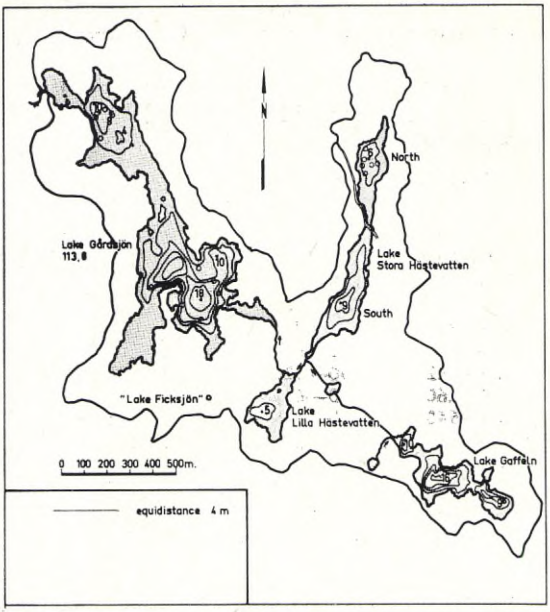


Fig. 1. Lake Gårdsjön watershed. Circles indicate sampling sites.

extent dependent on the acidification phase at the time of liming (cf HENRIKSON *et al.* 1984, in the press). Furthermore the biological community after liming may look different depending on the time passed since liming. Along a time scale the effects of liming can be separated into immediate, short term and long term effects. The immediate effects may be mainly caused by the rapid change of the abiotic environment. But the short and long term changes are probably more influenced by biotic interactions, such as predation and competition. In this paper we will elucidate immediate and short term changes of the crustacean zooplankton after liming of the acidified and fishless Lake Gårdsjön.

II. STUDY AREA

Lake Gårdsjön and Lake Stora Hästevatten belong to the same watershed (Fig. 1), approximately 50 km north of Göteborg on the Swedish west coast. The water runs from Lake Stora Hästevatten down to Lake Gårdsjön. Lake Stora Hästevatten is divided by a road bank with a drain which limits the water exchange between the northern and southern parts.

Table 1. *Morphometric data of Lake Gårdsjön and the* northern part of Lake Stora Hästevatten.

	Lake Gårdsjön	Lake Stora Hästevatten
Total drainage area (km^2)	2.1	0.18
Surface area (ha)	31	3.3
Volume ($10^6 \cdot m^3$)	1.5	0.10
Maximum depth (m)	18.5	6.5
Mean depth (m)	4.9	3.2

The bedrock consists mainly of gneiss and granite and is covered by a thin soil layer. Peatland and bare bedrock are also common. The vegetation within the area consists mainly of mixed coniferous forest and clearfellings.

Some morphological properties of the study lakes are presented in Table 1.

Our studies were performed in Lake Gårdsjön and the northern part of Lake Stora Hästevatten, which was used as control for the liming experiments in Lake Gårdsjön. All the lakes of the Lake Gårdsjön watershed were devoid of fish during the whole study.

Lake Gårdsjön was limed on April 26 and 27, 1982 by 110 metric tons of limestone (particle size 0—0.2 mm). Most of the limestone powder was spread in the littoral zone, but the deeper parts of the lake were also treated.

Some physical and chemical properties of water from the lakes are presented in Table 2. The whole Lake Gårdsjön watershed is treated in detail in ANDERSSON 1984 (in the press).

III. METHODS

The crustacean zooplankton was sampled six to seven times yearly at twelve sampling sites in Lake Gårdsjön and six sites in the northern part of Lake Stora Hästevatten (Fig. 1). The sampling sites were randomly selected from areas with depths greater than four metres according to the depth cart (Fig. 1). The sampling in Lake Gårdsjön was stratified into one southern and one northern part. In each part six sampling sites were chosen. The sampling was performed by vertical

Table 2. *Some physical and chemical parameters and the chlorophyll a content in Lake Gårdsjön and Lake Stora Hästevatten. Minimum and maximum epilimnic values from 17—20 sampling occasions per year.*

	Lake Gårdsjön		Lake Stora Hästevatten	
	before liming	after liming		
	1980—81	1982—83	1980—81	1982—83
pH	4.5—4.8	7.0—7.9	5.0—5.5	4.9—5.6
Alkalinity (meq · l^{-1})	0	0.20—0.54	0—0.01	0
Conductivity (mS · m^{-1})	5.7—7.2	7.5—10.5	4.8—6.2	5.5—6.7
Colour (mg Pt · l^{-1})	0—12	1—10	0—10	1—13
Secchi depth (m)	6.5—13.0	2.4—7.9	>6.5	>6.5
Total phosphorus (µg · l^{-1})	1—12	3—9	2—6	2—5
Nitrate (µg · l^{-1})	30—180	23—162	30—120	30—130
Ammonium (µg · l^{-1})	6—127	7—260	0—68	0—51
Total nitrogen (µg · l^{-1})	221—648	229—540	185—495	90—540
Calcium (mg · l^{-1})	1.6—2.4	7.3—13.7	2.0—3.7	2.2—2.6
Sulfate (mg · l^{-1})	7.8—13.0	8.5—11.2	6.7—11.4	8.8—10.3
Aluminium (mg · l^{-1})	0.10—0.49	0—0.48	0.01—0.45	0.03—0.77
Chlorophyll *a* (µg · l^{-1})	0—2.8	0.6—6.2	—	—

hauls with a net (mesh 65 µm, mouth area 324 cm^2, length 60 cm) from the bottom to the surface at a rate of approximately 0.5 m s^{-1}. All samples were fixed in 4 % formalin and were counted by subsample technique at 40 × magnification with an inverted microscope. The length of *Bosmina* was measured to the nearest 0.03 mm by an ocular micrometer on 10—70 randomly selected specimens from each sampling date.

Field experiments were performed in order to study the effects of *Chaoborus* predation on crustaceans. Ten cylindrical enclosures (diameter 0.96 m, length approx. 3 m) were placed in Lake Lilla Valeklintsvatten. This lake has a sparse population of salmonid fish, which did not allow the macroinvertebrate predators to establish detectable populations. Three of the enclosures were kept as controls while the others were stocked with *Chaoborus obscuripes* (V. D. WULP) and *Ch. flavicans* (MEIG.) of fourth instar in densities of approximately 700 ind m^{-2}. No crustacean prey were introduced to the enclosures, but natural populations of the lake were used. At the start of the experiment and after 8 and 20 days, zooplankton samples were taken by vertical net hauls (mesh 25 µm, mouth area 200 cm^2, length 0.5 m) within each enclosure. The length of *Bosmina* was measured by ocular micrometer to the nearest 0.01 mm on 50 randomly selected specimens from each sampling date.

Studies of the immediate effects of the lime treatment were done by analysing vertical hauls of 5.5 m each with a net of the same type as described in the first paragraph. Samples were taken at seven sites in the southern part of Lake Gårdsjön five days before and one day after the liming.

IV. RESULTS

The development of the most abundant zooplankton taxa in Lake Gårdsjön and Lake Stora Hästevatten during 1980—1983 is shown in Fig. 2 and Table 3. The most dominant taxa were the calanoid copepod *Eudiaptomus gracilis*, cyclopoid copepods (*Cyclops abyssorum* and others), the cladocerans *Bosmina* spp. (approx. 100 % *B. coregoni*), *Diaphanosoma brachyurum* and *Alonella nana*. The last mentioned species was not as abundant as the others but was regularly found at nearly all sampling sites. Several other species were also recorded, but only in low numbers at certain occasions and sites (Table 4). There was no great difference between the two lakes during acid conditions.

After liming of Lake Gårdsjön some changes, compared with previous years and the reference lake could be seen. An apparent difference was the lower number of *Bosmina* and higher number

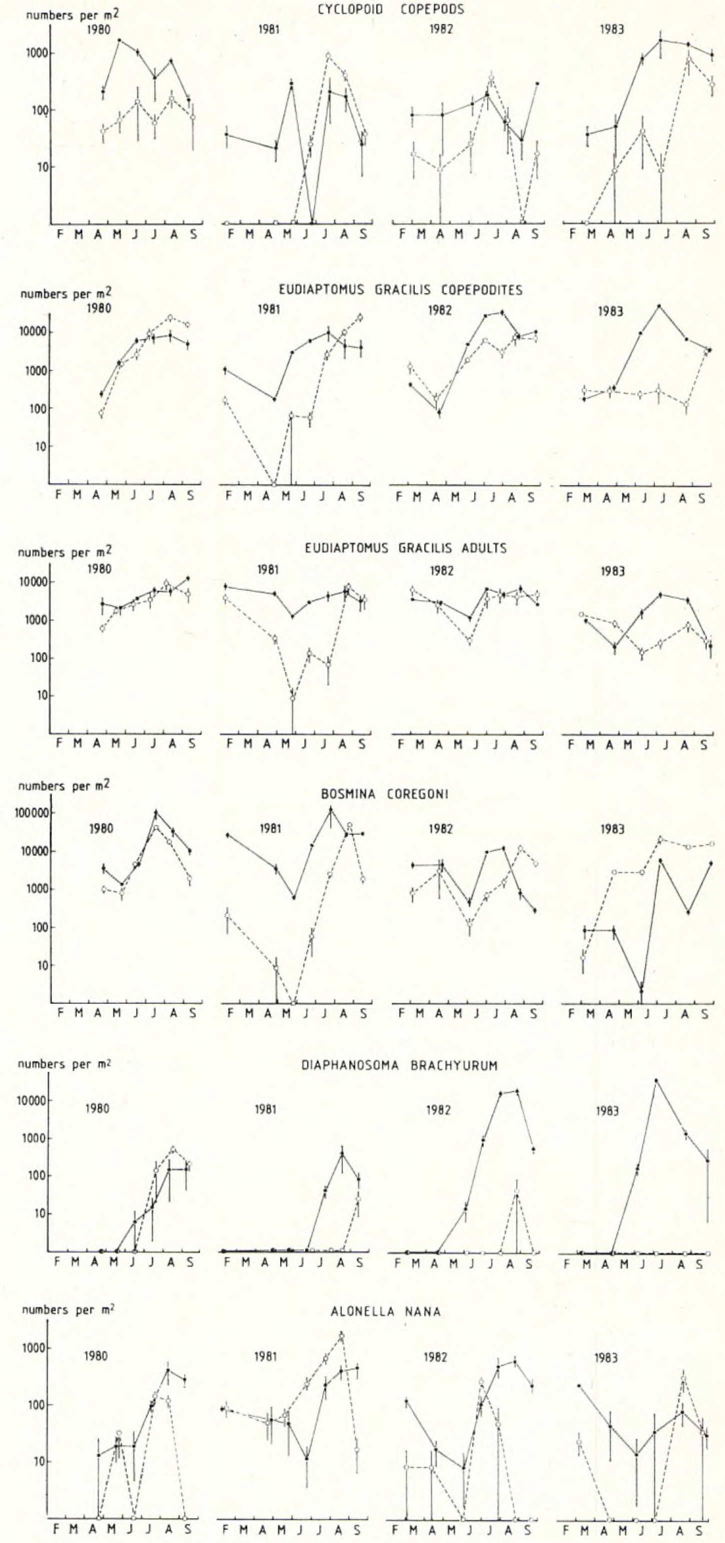


Fig. 2. Temporal development of the most abundant crustacean taxa in Lake Gårdsjön (solid line) and Lake Stora Hästevatten (broken line).

Table 3. *Number of crustaceans per sampling occasion in Lake Gårdsjön during 1980—83. Calculated from five sampling occasions comparable in time. Mean number ($\bar{x}$) per m² and standard error (SE).*

	1980		1981		1982		1983	
	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE
Calanoid copepods (adults + copepodites)	9 695	2 972	7 325	1 125	15 108	5 776	18 277	11 311
Cyclopoid copepods (adults + copepodites)	771	285	98	55	136	44	951	272
Nauplii	20 244	5 551	17 062	3 879	13 942	3 179	13 271	3 992
Bosmina spp	10 646	5 953	15 815	6 310	3 105	1 795	2 261	1 325
Diaphanosoma brachyurum	62	36	98	77	4 314	3 906	8 658	8 135
Alonella nana	150	84	195	96	193	109	43	12
Other cladoceran species	17	7	31	15	35	29	12	7

of *Diaphanosoma* (Fig. 2). No great changes in number of species occurred during the investigation period (Table 4).

The results of the study of immediate effects of lime application are summarized in Table 5. There were no significant ($p > 0.05$, Mann-Whithney U-test) changes in the numeric relations between different taxonomical groups.

The measurements of the *Bosmina* body size are summarized in Fig. 3. A specific pattern in the size development could be seen with the largest individuals occurring early in the year and thereafter a decrease down to the smallest individuals in summer. The development was similar in both lakes and during all three years, 1981—83, and hence was not changed after liming.

Table 4. *Recorded crustacean taxa in Lake Gårdsjön and Lake Stora Hästevatten. Percentage samples with occurrence.*

	Lake Gårdsjön		Lake Stora Hästevatten	
	before liming	after liming		
	1980—81	1982—83	1980—81	1982—83
Total number of samples	156	132	77	78
PELAGIC TAXA				
Eudiaptomus gracilis G. O. Sars	97	98	95	96
Cyclopoida *	65	63	87	55
Bosmina coregoni Baird	98	80	78	91
Diaphanosoma brachyurum Liéven	24	67	21	1
Ceriodaphnia quadrangula (O. F. Müller)	13	7	1	0
BENTHIC AND LITTORAL TAXA				
Alonella nana (Baird)	59	64	62	23
A. excisa (Fischer)/*exigua* (Lilljeborg)	0	1	3	0
Alona spp *	14	7	9	8
Eurycercus lamellatus (O. F. Müller)	0	3	0	0
Streblocerus serricaudatus (Fischer)	0	1	8	6
Acantholeberis curvirostris (O. F. Müller)	1	0	1	3
Acroperus harpae Baird	2	2	3	0
Chydorus spp *	4	2	0	0
Ilyocryptus sordidus (Liéven)	1	3	0	0
Macrothrix sp	0	0	1	0
Polyphemus pediculus (L.)	0	2	0	0
Sida crystallina (O. F. Müller)	1	2	1	0

* Among these taxa the following species were noted: *Cyclops abyssorum* G. O. Sars, *Alona affinis* Leydig, *A. costata* (Sars), *A. rustica* T. Scott, *A. quttata* Sars, *Chydorus sphaericus* (O. F. Müller), *C. piger* Sars.

Table 5. *Crustacean zooplankton at seven sampling sites before (April 21) and after (April 28) lime treatment of Lake Gårdsjön on April 26 and 27, 1982. Mean number per haul ($\bar{x}$) and standard error (SE).*

	Before liming		After liming	
	$\bar{x}$	SE	$\bar{x}$	SE
Eudiaptomus gracilis (adults+copepodites)	72	11	69	6
Cyclopoids (adults+copepodites)	4	2	3	2
Nauplii	260	24	290	22
Bosmina coregoni	27	9	26	5
Alonella nana	7	2	30	5

The results of the field enclosures experiments are summarized in Table 6. In the enclosures with *Chaoborus flavicans* a significant ($p < 0.05$, t-statistics) decrease of the *Bosmina* length was noted after eight experimental days. In the control enclosure no significant ($p > 0.05$) reduction was recorded. The *Bosmina* abundance in the test enclosure was also significantly ($p < 0.05$, t-statistics) reduced during the first eight days.

V. DISCUSSION

The changes of the fauna in lakes during acidification have been attributed to different origins, (a) abiotic, such as a toxic impact of acid water and or other toxic elements in the acid water, and (b) biotic, such as altered predator-prey relations (ERIKSSON *et al.* 1980). These two main types of mechanisms are also involved in the fauna changes following a lime treatment.

Before the liming of Lake Gårdsjön we posed some questions about the development of the crustacean zooplankton community. These were: (1) Does the rapid change of the abiotic environ-

Table 6. *Enclosure experiment with Chaoborus spp and Bosmina coregoni. Three controls and seven experimental enclosures were used. Mean values ($\bar{x}$), standard error (SE) and number of Bosmina measured (n).*

Date	Abundance (number per net haul)				Body length (mm)					
	control		experiment		control			experiment		
	$\bar{x}$	SE	$\bar{x}$	SE	$\bar{x}$	SE	n	$\bar{x}$	SE	n
May 7	58	13	63	10	0.46	0.01	100	0.46	0.01	100
May 15	55	14	0.20	0.06	0.45	0.01	89	0.49	0.02	31
May 26	26	6.8	0.29	0.08	0.44	0.01	71	0.39	0.01	51

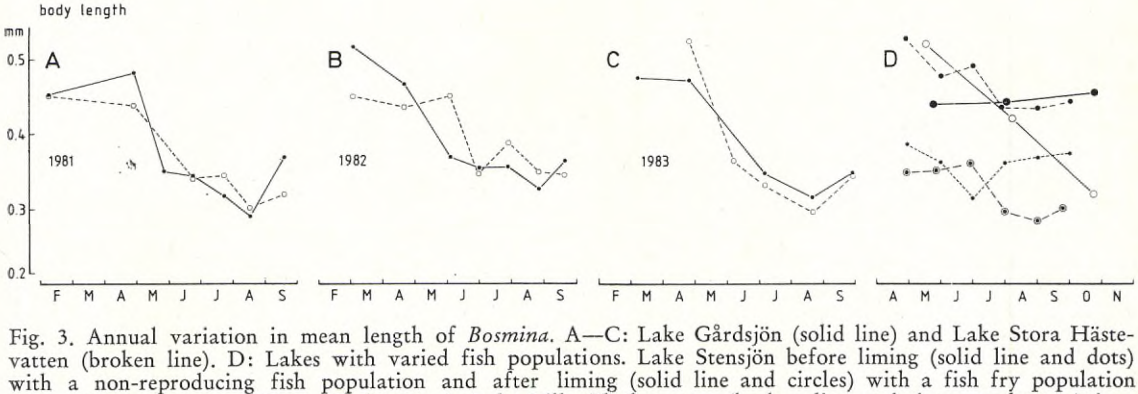


Fig. 3. Annual variation in mean length of *Bosmina*. A—C: Lake Gårdsjön (solid line) and Lake Stora Hästevatten (broken line). D: Lakes with varied fish populations. Lake Stensjön before liming (solid line and dots) with a non-reproducing fish population and after liming (solid line and circles) with a fish fry population (after HENRIKSON *et al.* 1984, in the press). Lake Lilla Blackevatten (broken line and dots, at the top) is a non-acidified lake and has a sparse fish population, Lake Lilla Stockelidsvatten (broken line and dots in circles) and Lake Lilla Mörtevatten (broken line and dots are non-acidified lakes with dense fish populations (after STENSON 1976).

ment give negative immediate effects? (2) Do species, sensitive to the acid environment increase or, if they were eliminated during acidification, reestablish their populations? (3) If phytoplankton production and biomass increase will the zooplankton abundance and biomass also increase? (4) If zooplankton predators increase will the prey populations be affected? (5) Will the structure of the zooplankton community on the whole be the same after liming as during acid conditions?

Regarding the first question there were great and rapid changes of the environment. For example the pH value raised from around 4.6 to 7.6 during just a few days, *i.e.* the acidity was reduced thousandfold. Other physical and chemical properties were also greatly changed (Table 2). But according to our zooplankton samples just before and after liming, no negative effects could be discovered. There were no significant changes in the abundance of *Eudiaptomus gracilis*, cyclopoid copepods or *Bosmina* spp (Table 4) and no other signs of negative influence were noted. In fact, the only change was an increase of *Alonella nana.* Hence, we did not detect any lethal influence on species at the time of liming. In relation to this we can mention that negative effects were noted on phytoplankton shortly after the liming (LARSSON, unpubl. data).

Some species, abundant in non-acidified oligothrophic lakes are only rarely found in acidified lakes. Such species are *Holopedium gibberum* Zaddach and *Daphnia cristata* G. O. SARS and the absence of these two cladocerans has been said to be caused by toxic components or altered predator-prey relations (STENSON and OSCARSON 1984, in the press). If the acid environment is the cause for the absence of the two species they should appear after liming. But still two years after the lime treatment we have not recorded a single specimen of either *Holopedium gibberum* or *Daphnia cristata.* Some "new" species have been recorded after liming but only on very few occasions (Table 4). Thus, the answer to our second question seems to be no. It may not, however, be possible to reject the hypotheses because the effects of improved water quality may be masked by the effect of invertebrate predation which has not changed structurally but probably became more intensive after liming. The dominant predators are still invertebrates like *Chaoborus* spp and corixids. The impact of invertebrate predation, especially from *Chaoborus* is well documented (*e.g.* KERFOOT 1980). Thus, the invertebrates may suppress or prevent immigration of species like *Holopedium gibberum* and *Daphnia cristata* and not until the predators are eliminated, by fish introduction, will the two cladocerans establish detectable populations. This development took place in the non-acidified Lake Lilla Stocklidsvatten where the fish population was eliminated experimentally. After that, *Chaoborus* spp and the corixid species *Glaenocorisa propinqua* bloomed up while *Holopedium gibberum* and *Daphnia cristata* disappeared (STENSON *et al.* 1978). After restocking of fish the invertebrate predators were reduced by predation down to undetectable populations and the *Holopedium gibberum* and *Daphnia cristata* returned. A similar development was demonstrated by ERIKSSON *et al.* (1983) in the liming of lakes devoid of fish. Not until fish were introduced and had eliminated *Chaoborus* and *Glaenocorisa* were *Holopedium* and *Daphnia* recorded. Our and other studies therefore strongly indicate that the absence of certain cladoceran species in lakes devoid of fish after liming may be caused by macroinvertebrate predators. But, of course, the dispersal possibilities may be of great importance in the return of a species. *Holopedium* is, in fact, found in a small bog pool, "Lake Ficksjön", in the Lake Gårdsjön watershed (Fig. 1), but it has no direct water connection with Lake Gårdsjön. In fact, *Holopedium* from Lake Ficksjön can survive and reproduce in Lake Gårdsjön according to preliminary field exepriments. So, if dispersal is an obstacle to the establishment of certain species after liming, our short term studies must be prolonged in order to detect this.

In our third question we asked, if phytoplankton was favoured by the liming would not the zooplankton abundance and biomass increase? After the immediate shock effects the phytoplankton recovered and reached production and biomass levels far above those during acid conditions. Furthermore the species number increased considerably (LARSSON, unpubl. data). Hence, conditions necessary for an increased zooplankton abundance and biomass seemed to be present. The total abundance of the crustaceans did not, according to our semi-quantitative data, show any increase. We have not measured the biomass but a rough estimation of the biomass (based on literature weights used by NYBERG (1984)) indicates an increase.

Regarding the fourth question, *i.e.* would increased predator populations affect prey populations, a significant increase in numbers was noted for the predatory groups *Chaoborus* spp, corixids and dytiscids in Lake Gårdsjön after liming (HENRIKSON and OSCARSON 1984). The population density of *Chaoborus* spp and corixids increased by a factor of 6—7 and 2—3 respectively. Judging from the abundance, *Chaoborus flavicans* and the corixid *Glaenocorisa propinqua* ought to be the most important predators in the pelagical zone. The cause of the increased predator populations is difficult to pin point but there are some alternative explanations. Firstly, if the crustacean zooplankton production increased this would give an opportunity for a correspondingly increased abundance on higher trophic levels. Secondly, other prey populations than crustaceans, *e.g.* rotifers, may be important. The rotifers in Lake Gårdsjön increased in abundance and species number after the liming (HENRIKSON *et al.* unpubl.). This may lead to an improved food situation for the predators or possibly just for certain early stages of their life cycles. Increased availability of rotifers for early instars of *Chaoborus* may imply a "break through the bottleneck" (NEILL and PEACOCK, 1980). This means that an improved food level for a sensitive early instar may lead to increased survival and larger populations of all instars.

Obvious changes in the prey crustacean populations after liming were the decrease of *Bosmina* and the increase of *Diaphanosoma brachyurum* (Fig. 2 and Table 3). That these changes were consequences of the liming are strengthened by opposite changes in the control lake. There, in fact, *Bosmina* increased and *Diaphanosoma* decreased their population density (Fig. 2). The decrease of *Bosmina* populations is probably an effect of the increased predatory pressure. This statement is based on our enclosure experiment studies and on other studies. In our enclosure experiments we found that *Chaoborus* spp were able to significantly reduce the abundance of natural *Bosmina* populations within eight days (Table 5). Other studies have also shown that *Chaoborus* severely affected the *Bosmina* abundance (*e.g.* NEILL, 1981). NYBERG (1984), who also studied *Chaoborus* and crustacean zooplankton interactions in acidified and limed lakes, states that it is possible that *Chaoborus* is able to eradicate herbivore crustaceans in lakes devoid of fish. Regarding the corixids NYMAN *et al.* (in prep.) in enclosure experiments, similar to our *Chaoborus* experiments, showed that *Glaenocorisa propinqua* significantly reduced the *Bosmina* abundance. Hence, there are several experimental studies supporting the conclusion that the decrease of *Bosmina* in Lake Gårdsjön after liming is caused by the increased invertebrate predation. Another way to detect effects of predation on *Bosmina* is to study the body size. For example fish have been shown to affect *Bosmina* resulting in smaller individuals as fish are visually dependent predators selecting the larger prey before the smaller. Therefore, the *Bosmina* mean body size is smaller in lakes with high fish predation than in lakes with low predation (STENSON 1976). In this study we have measured the mean length of *Bosmina* in our fishless lake at different sampling occasions before and after the liming. The mean body size had a similar annual variation in both Lake Gårdsjön and Lake Stora Hästevatten and the cycle was not changed after liming. The greatest mean length was noted in February—March after which it declined up to 45 % to a minimum value during summer and thereafter it increased (Fig. 3). This range in variation makes our lakes different from lakes where fish are present. In those lakes there is a much smaller reduction of the mean length during the spring and summer (Fig. 3) and the minimum values are dependent on the intensity of fish predation, *i.e.* the higher predation the smaller the *Bosmina* individuals. As fish predators are able to reduce the mean size of *Bosmina* it may be the invertebrates that reduce the mean size in lakes devoid of fish. This hypothesis was tested in field enclosure experiments on natural *Bosmina* populations in a lake of very low fish predation intensity and devoid of *Chaoborus* and corixids (because of presence of fish). *Chaoborus flavicans*, introduced in densities approximately those in acid lake, did reduce the mean body size of *Bosmina* significantly ($p < 0.05$, t-statistics) (Table 5). NYMAN *et al.* (in prep.) made a similar experiment and noted a tendency of decreasing mean size of *Bosmina* exposed to the predator *Glaenocorisa propinqua*. These experiments indicate that the invertebrate predators, at least partly, may be responsible for the *Bosmina* size decline. NEILL (1981) did not find any statistically significant reduction (but a consistent trend) of the *Bosmina* mean size after exposure to two different densities of *Chaoborus* in enclosure experiments. There are however, differences between our and NEILL's experiments. NEILL used another species, *Bosmina longirostris*, taken from a lake devoid of fish, *i.e.* probably already influenced by *Chaoborus*. To summarize, in our lakes devoid of fish *Bosmina* size varied annually in a different way compared to populations in lakes with fish populations.

Another cladoceran species shown to be susceptible to *Chaoborus* predation is *Diaphanosoma brachyurum* (*e.g.* FEDERENKO 1975). In Lake Gårdsjön *Diaphanosoma* increased considerably after liming (Fig. 2) despite the coincidental increase of *Chaoborus*. Hence, the increased potential predation must be compensated by some other change that favours *Diaphanosoma*. It can probably not be better water quality, measured for example as pH and total aluminium content, because high densities of *Diaphanosoma* are found in the southern part of Lake Stora Hästevatten (Fig. 1) (STENSON and OSCARSON 1984, in the press), where pH is just above 5 and aluminium content around 170 µg/1, and in other acid lakes (HULTBERG and ANDERSSON 1981, ERIKSSON *et al.* 1983, HENRIKSON *et al.* 1984, in the press). The great changes in the phytoplankton may contain the explanation for the *Diaphanosoma* explosion. The composition of the phytoplankton may be of importance as *Diaphanosoma* established a dense population in the southern part of Lake Stora Hästevatten but only a sparse population in Lake Gårdsjön (before liming), while the phytoplankton flora was somewhat different in these lakes (LARSSON unpubl. data). Another possible cause may be competition as both *Bosmina* and *Diaphanosoma* feed on small particles. A decline in number of one of the competitors would lead to an increase to the other, with the food level unchanged. The development in Lake Gårdsjön may be an example of this. However, one crucial question remains about the reason for the decline of *Bosmina*. This problem may be explained in terms of predation by the larger *Chaoborus* population. *Chaoborus* is an ambush predator waiting for the prey to come within the attack distance. Fast moving prey organisms will have a higher encounter rate and therefore be more vulnerable to *Chaoborus* predation. In fact, *Bosmina* is a faster swimmer than *Diaphanosoma* (4 times as fast), and faster than the other potential prey organisms as well (GERRITSEN 1980). This implies that *Bosmina* is more often caught by *Chaoborus*, which favours *Diaphanosoma* when they are competing for common resources.

In conclusion, the answer to the fourth question is that the predation from increased predator populations probably has caused the decline of *Bosmina* populations, resulting in decreased competition which favours *Diaphanosoma*.

Regarding the fifth question the structure of the zooplankton community seemed largely to be the same after liming. Before liming the community was quantitatively dominated by *Eudiaptomus gracilis*, *Bosmina coregoni*, lower numbers of cyclopoid copepods and *Alonella nana* and several other rarely occurring species. This structure was the same after liming, but *Bosmina* had partly been replaced by *Diaphanosoma brachyurum*.

To summarize our lime effects; we found no detectable immediate shock effects and the short term development showed changes in the proportions of existing species but no "new" species were established. Our studies describe only short term effects and we may just have seen one step of a succession. This was indicated in the development of *Bosmina* and *Diaphanosoma*. The former species shows a continuous decline in abundance while the latter shows an opposite development and thus the situation is not yet stable. Studies of the long term effects of liming may therefore give other results than those presented here.

Few other studies of the lime effects on crustacean zooplankton fauna in lakes devoid of fish have been published. ERIKSSON *et al.* (1983), however, presented studies from several lakes before and after liming. The abundance of most observed species increased after liming. Among these species *Diaphanosoma*, as in Lake Gårdsjön, showed a great increase. HULTBERG and ANDERSSON (1981) reported increased biomass and new cladoceran species after liming and fish introduction. But when comparing the lime effects one must be aware of the faunal structure in the lakes before liming, a point which has been stressed by HENRIKSON *et al.* (1984, in the press). Lake Gårdsjön was a typical phase 3 lake with no fish and with invertebrates as top predators. After liming of a phase 3 lake the invertebrate predator system will still dominate (if fish are not introduced) and hence no radical changes may be expected in communities like zooplankton that are greatly influenced by predation. Liming a phase 2 lake (with a non-reproducing fish stock) gives another outcome. In Lake Stensjön a dramatic decrease in abundance of almost all crustaceans occurred probably due to intensive predation from fish fry, which became abundant due to the better water quality (*op. cit.*). The predation impact of fish fry was also indicated in reduction of the mean size of *Bosmina coregoni* (Fig. 3) and *Holopedium*.

The importance of the predator system in lakes is also demonstrated by ERIKSSON *et al.* (1983). Lake Blanksjön, Lake Iglafallssjön and Lake Vibollsjön, all typical phase 3 lakes, were limed without fish introduction. An increased crustacean abundance was noted but no enrichment of species. Almost two years after liming, two lakes were stocked with fish fry and yearlings which eliminated the invertebrate predators. Two years after the fish introduction cladocerans like *Holopedium* and *Daphnia longispina* appeared and the copepod dominance was exchanged for cladoceran dominance. The third lake, still devoid of fish, was dominated by copepods. Hence, if we are going to predict the biological outcome of lime treatment of a specific lake, we must know the community structure of both the prey and the predators during acid conditions.

VI. FINAL REMARKS

Although several abiotic properties of Lake Gårdsjön have been restored to pre-acid conditions by the lime treatment, the zooplankton structure has not become the same as in normal oligothrophic lakes. This is mainly because the "normal" top predators, *i.e.* fish, do not occur there. Fish seem to be key organisms in the development during both acidification and liming. Therefore in order to restore acidified lakes it is not sufficiant to only treat them with lime. Important organisms must be introduced. The best way to minimize negative biological effects is to lime lakes during phase 1 in order to improve the buffering capacity before any great biological damage has taken place.

VII. ACKNOWLEDGMENTS

This study was carried out within the integrated Lake Gårdsjön project. This work was financially supported by grants from the Swedish National Environmental Protection Board and the Swedish Natural Science Research Council.

VIII. REFERENCES

ANDERSSON, F. and B. OLSSON. 1984. Lake Gårdsjön — an acid forest lake and its catchment. Ecol. Bull. (Stockholm) 37. (In the press.)

ERIKSSON, F., E. HÖRNSTRÖM, P. MOSSBERG and P. NYBERG. 1983. Ecological effects of lime treatment of acidified lakes and rivers in Sweden. Hydrobiologia 101: 145—164.

ERIKSSON, M. O. G., L. HENRIKSON, B.-I. NILSSON, H. G. NYMAN, H. G. OSCARSON, J. A. E. STENSON and P. K. LARSSON. 1980. Predator-prey relations, important for the biotic changes in acidified lakes. Ambio 9: 248—249.

FEDORENKO, A. Y. 1975. Feeding characteristics and predation impact of *Chaoborus* (Diptera, Chaoboridae) larvae in a small lake. Limnol. Oceanogr. 20: 250—258.

GERRITSEN, J. 1980. Adaptive responses to encounter problems. p. 52—62. *In* Evolution and ecology of zooplankton communities. Ed.: W. C. Keerfoot. Special symposium 3, Amer. Soc. Limnol. Oceanogr. University Press of New England, Hanover. New Hampshire and London, England.

HENRIKSON, L. and H. G. OSCARSON. 1978. Fish predation limiting the abundance and distribution of *Glaenocorisa p. propinqua* (Hemiptera). Oikos 31: 102—105.

— and H. G. OSCARSON. 1984. Lime influence on macro-invertebrate zooplankton predators. Rep. Inst. Freshw. Res., Drottningholm 61: 93—103.

— H. G. NYMAN, H. G. OSCARSON and J. A. E. STENSON. 1984. Changes in the zooplankton community after lime treatment of an acidified lake. Verh. internat. Verein. Limnol. 22. (In the press.)

HULTBERG, H. and I. B. ANDERSSON. 1981. Liming of acidified lakes: induced long-term changes. Wat., Air Soil Poll. 18: 311—331.

JOHANSSON, K. and P. NYBERG. 1981. Acidification of surface waters in Sweden — effects and extent

1980. Inform. Inst. Freshw. Res., Drottningholm (66). 118 p. (In Swedish with English summary.)

Keerfoot, W. C. (Ed.) 1980. Evolution and ecology of zooplankton communities. Special symposium 3, Amer. Soc. Limnol. Oceanogr. University Press of new England, Hanover. New Hampshire and London, England. 793 p.

Neill, W. E. 1981. Impact of *Chaoborus* predation upon structure and dynamics of a crustacean zooplankton community. Oecologia 48: 164—177.

— and A. Peacock. 1980. Breaking the bottleneck: Interactions of invertebrate predators and nutrients in oligotrophic lakes. p. 715—724. *In* Evolution and ecology of zooplankton communities. Ed.: W. C. Keerfoot. Special symposium 3, Amer. Soc. Limnol. Oceanogr. University Press of New England, Hanover. New Hampshire and London, England.

Nyberg, P. 1984. Impact of *Chaoborus* predation on planktic crustacean communities in some acidified and limed lakes in Sweden. Rep. Inst. Freshw. Res., Drottningholm 61: 154—166.

Raddum, G. G. and A. Fjellheim. 1984. Acidification and early warning organisms in freshwater in western Norway. Verh. internat. Verein. Limnol. 22: 1973—1980.

Stenson, J. A. E. 1976. Significance of predator influence on composition of *Bosmina* spp. populations. Limnol. Oceanogr. 21: 814—822.

— 1978. Differential predation by fish on two species of *Chaoborus* (Diptera, Chaoboridae). Oikos 31: 98—101.

— T. Bohlin, L. Henrikson, B.I. Nilsson, H. G. Nyman, H. G. Oscarson and P. Larsson. 1978. Effects of fish removal from a small lake. Verh. internat. Verein. Limnol. 20: 794—801.

— and H. G. Oscarson. 1984. Crustacean zooplankton in the acidified Lake Gårdsjön system. Ecol. Bull. (Stockholm) 37. (In the press.)

Zaret, T. M. 1980. Predation and freshwater communities. Yale University Press, New Haven and London. 187 p.

Effects of pH and Different Levels of Aluminium on Lake Plankton in the Swedish West Coast Area

EINAR HÖRNSTRÖM, CHRISTINA EKSTRÖM and M. OSAMA DURAINI

National Swedish Environment Protection Board, P. O. Box 1302, S-171 25 Solna, Sweden

ABSTRACT

Based upon the results from regional investigations in lakes of the Swedish west coast area between 1973 and 1983 and from biotest experiments, the relationship between the composition of plankton and pH, nutrient levels and the concentration of aluminium, is examined.

The conspicuous absence of several plankton species in acid lakes is not caused by the low pH-value as such, but rather by a raised aluminium supply from the surrounding environment, which produces oligotrophic waters through precipitation of phosphorus. A comparison between plankton communities in strongly acid lakes with low and high levels of aluminium respectively, indicates, however, that aluminium also through its pure toxic effects contributes to the deficiency of plankton species. This is supported by the results from biotests, showing reduced growth of many phytoplankton species at a concentration of 100 μg Al/l, and the inhibited growth of 13 out of 19 tested species at 200 μg Al/l. Among those were desmids and diatoms. At the same aluminium concentrations morphological damages were observed, *e.g.* cell enlargement, destruction of cell constituents and other developmental defects as a result of disturbances in the reproduction process.

Most zooplankton species are apparently not sensitive to low pH-values as such but react to the oligotrophication process.

In lakes where the aluminium level exceeded 180 μg/l, 10 species out of ca 30 totally registered were lacking, which partly may be a result of aluminium toxicity since the introductory tests with *Daphnia magna* and *Acroperus harpae* showed that Al-concentrations of 150—300 μg/l are critical.

I. INTRODUCTION

The deficiency of plankton species is one of the most important consequences of the acidification of lake water (ALMER *et al.* 1978, NRCC 1981). This partly is an effect of acidity as such. The reduction of the nutrient supplies is, however, a considerably more important factor. The plankton diversity has a connection to the trophic level of the lakes, and when the supply of phosphorus diminishes as an effect of the rapidly increased mobilization of aluminium at pH-values lower than 5.5, (ALMER *et al.* 1978), the number of phytoplankton species in particular is drastically reduced. Several species which generally are lacking in the most acid lakes, are frequently recorded in extremely oligotrophic waters at pH 5.3—6.5. These species actually occur accidentally also in a few strongly acid lakes during special circumstances (see below), and thus they can withstand both oligotrophy and low pH-values. Considering these facts, evidently other environmental mechanisms are of importance.

Increased concentrations of different metals, more or less toxic, is a secondary effect of the acidification of natural water systems (ALMER *et al.* 1978). Attention should therefore be drawn to the metal toxicity on plankton.

Results from our introductory field- and laboratory algal tests on sensitivity to manganese, zinc, copper and cadmium, have shown that these metals are toxic mainly at pH-values above 6, and that toxicity was recorded only at concentrations higher than those registered in our acid lakes. These metals therefore seem to be of less importance in this connection. Aluminium is, in contrast to the other metals, more toxic to phytoplankton at pH 5.8—5.1, that is the pH-interval where the level of aluminium is rapidly increasing in water subjected to acidification. Consequently the relation between aluminium concentrations and

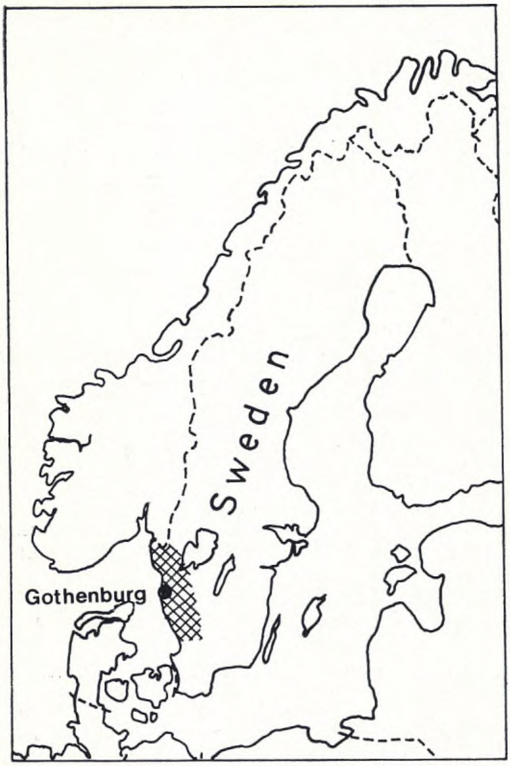


Fig. 1. Location of the investigated lakes.

the distribution of plankton species deserves special interest.

This paper represents an attempt to separate and explain the relative importance of low pH, nutrient depletion and aluminium toxicity to plankton in acidified waters.

The presentation is in part a summary of a paper in Swedish (Hörnström and Ekström 1983) containing more detailed primary information (species lists and results from chemical and physical analyses).

II. MATERIAL AND METHODS

Data on plankton distribution in more or less acidified lakes were available from an inventory covering 65 lakes, evenly distributed over the Swedish west coast area (Fig. 1). The investigation was made in August 1976. Between 1973 and 1983 further information was obtained by closer investigations (3 to 10 times a year) of some of these lakes, situated in the district of Gothenburg.

Samples were taken in the middle of the lakes, phytoplankton mostly at 0.5 m depth and zooplankton by plankton net (mesh size 25 and 75 μm) sampling (13—0 metres). The samples were treated according to Willén (1974).

Phytoplankton was examined according to the Utermöhl-technique (Willén 1974). The total sum of species in each sample (100 ml) was determined and the frequency of each species was estimated. The trophic index of the lake was calculated by means of trophically indicative phytoplankton species and their frequency (Hörnström 1981). Zooplankton was qualitatively analysed with the estimation of their relative abundance.

Biotest experiments were performed in water from the ultraoligotrophic Lakes Östra Nedsjön (pH ca 6), Tvärsjön (pH ca 4.7) and Härsevatten (pH ca 4.4). The water was taken to the laboratory in plastic cans, and was kept deep frozen until the experiments were performed. Prior to the tests the water was filtered through a membrane filter (1.2 μm) and 5 μg of PO_4-P per litre was then supplied.

Plankton species used in the biotests were isolated in August 1983 from lakes, having pH-values about 7.

At the beginning of the phytoplankton experiments ca 500 cells/ml of algal suspension was added to 100 ml lake water in 250 ml E-flasks. Each experiment was performed in triplicate samples. All the experimental vessels were placed on agitators illuminated by flourescence lamps (Philips 40 W) giving an irradiance of 2000 lux. The illumination period was 12 h/day. After 3—4 days of growth the increased cell number or the relative *in vivo* chlorophyll *a* concentrations was determined by either using the Utermöhl-technique or a fluorometric technique (Turner 111).

In the zooplankton biotests, *Daphnia magna* was used for the acute toxicity test, performed in water from Lake Östra Nedsjön at pH 5.5, filtered through a membrane filter (0.4 μm). 20 animals of the age of 4—5 days were transferred into beakers, each containing 20 ml water. Four parallel beakers without aluminium were used as controls and two for each aluminium concentration.

Acroperus harpae was tested in one-litre aquariums containing filtered lake water at pH 5.5. The duration of this experiment was 5 weeks at which the plankton was fed on algal and bacterial suspension.

The stock solutions of aluminium used in all biotests were prepared from $Al_2(SO_4)_3$ 18 H_2O. Test solutions were prepared and added to the vessels at the start of the experiments. pH was adjusted by the addition of Na_2CO_3 and HCl, respectively, when required.

pH and concentrations of phosphorus and aluminium were analysed according to the described Swedish methods (Röndell 1982).

III. RESULTS

Phytoplankton

About 20 species of phytoplankton occurred during August in the strongly acid (pH < 5.3) lakes compared to about 50 species in lakes at pH-values above 6. Motile flagellates were the only phytoplankton present in the most oligotrophic acid lakes, *e.g. Peridinium inconspicum, Amphidinium* sp, *Gymnodinium uberrimum* and *Dinobryon pediforme.*

Low pH generally occurred at the same time as nutrient depletion and high levels of toxic metals, which could have been important to the species composition. Chemical and physical analyses, however, showed that the concentrations of nutrients and metals, with the exception of calcium, were similar at pH-values above ca 5.7 (Hörnström and Ekström 1983). The phytoplankton composition in these lakes at pH 5.8—6.3 differed from that in lakes at higher pH-values (including limed lakes). 18 species out of ca 130 totally registered were lacking in lakes of the lower pH-range (Table 1).

There were generally very low levels of total phosphorus in the acid lakes (< 2 µg/l). In a few waters, exhibiting a relatively rich phosphorus supply (3—10 µg/l), however, totally 50—60 phytoplankton species were recorded in addition to the ones normally occurring during extreme oligotrophy. These species, *e.g.* cyanophyceans, diatoms and desmids (Table 2), which were generally not recorded in acid lakes, are not sensitive to low pH alone.

Table 1. *Phytoplankton species registered in lakes with pH-values above 6.3 only (August 1976).*

Cyanophyceae
Aphanocapsa elachista
Aphanothece ellipsoidea
Gomphosphaeria lacustris

Chlorophyceae
Gyromitus cordiformis
Planctosphaera gelatinosa
Quadrigula pfitzerii
Scenedesmus acuminatus
,, *denticulatus*
Spondylosium planum

Chrysophyceae
Pseudokephyrion poculum
Stichogloea doederleinii

Bacillariophyceae
Cyclotella comta
Synedra nana

Dinophyceae
Ceratium hirundinella
Gymnodinium fuscum
,, *helveticum*
,, spp.
Peridinium willei

The growth rate of 15 species, not recorded in the strongly acid lakes, was studied. Their growth rate distinctively varied (Table 3). Especially *Oscillatoria, Synedra* and *Nitschia,* but also *Scenedesmus,* had highly reduced growth rate at low pH, while 7 species, including the desmids *Cosmarium* and *Staurastrum,* grew faster at pH 4.8 or 5.4 than at pH 6.0. The growth of *Crucigenia, Dictyosphaerium* and *Eudorina* may be independent of pH, but thin cultures combined with a high variance between the replicates, made this uncertain.

In Lake Ömmern the pH-value decreased from about 6 in 1976 to 5.3 in 1981. During the same period the concentration of aluminium in the water increased from 10 to about 100 µg/l. In connection with this the level of total phosphorus decreased from 5 to 2 µg/l (Fig. 2). In the unacidified (pH ca 6.5) adjacent Lakes Västra Nedsjön and St Färgen with low levels of aluminium (ca 10 µg/l) the simultaneous development was different. The concentration of phosphorus

Table 2. *Phytoplankton species registered in acid (pH < 5.3) lakes at especially rich nutrient supply, in humic (colour > 30) lakes (H) or in waters with low levels (about 50 µg/l) of aluminium (x). (August 1976).*

Cyanophyceae	
Anabaena flos-aquae	x
Chroococcus limneticus	x
Merismopedia glauca	x
,, *tenuissima*	x
Chlorophyceae	
Botryococcus braunii	
Chlorogonium sp	
Elakatothrix genevensis	
Koliella longiseta	x
,, sp	x
Monoraphidium dybowskii	
Oocystis sp	
Pediastrum tetras	x
Scenedesmus ecornis	x
Scourfieldia complanata	H
,, sp	H
Sphaerocystis schroeterii	x
Tetraedron minimum	H
Tetrastrum komarekii	
Willea irregularis	
Desmidiales	
Arthrodesmus octocornis	H
Staurastrum anatinum	x
,, *pseudopelagicum*	x
Staurodesmus extensus	H
,, *glaber v. limnophilus*	H
Chrysophyceae	
Bicosoeca spp	H
Bitrichia chodatii	
,, *ollula*	
Chromulina spp	
Chrysococcus sp	H
Chrysolycos planctonicus	
Dinobryon bavaricum	H
,, *borgei*	H
,, *pediforme*	
,, *suecicum*	H
Kephyrion boreale	
Mallomonas caudata	
Paramastix conifera	
Pseudokephyrion entzii	
,, *gibbosum*	
Prymnesiaceae spp	
Spiniferomonas spp	
Uroglena spp	
Diatomeae	
Asterionella formosa	x
,, *gracillima*	H
Rhizosolenia eriensis	H
Tabellaria fenestrata	
,, *flocculosa*	
Cryptophyceae	
Cryptaulax spp	
Cryptomonas spp	
Cyatomonas truncata	H
Rhodomonas minuta	
Chloromonadophyceae	
Gonyostomum semen	H

Table 3. *Relative growth rate of 15 species of phytoplankton at different pH-values (4.8, 5.4 and 6.0), where 100 represents the fastest growth rate, and the other figures percentage shares of that. (+ = thin cultures or high variance).*

Table 3 continued.

pH	4.8	5.4	6.0
Cyanophyceae			
Oscillatoria limnetica fa	5	61	100
Chlorophyceae			
Chlamydomonas sp	100	15	22
Crucigenia tetrapedia	+	+	+
Dictyosphaerium elegans	+	+	+
Eudorina elegans	+	+	+
Monoraphidium dybowskii	100	100	100
Monoraphidium griffithii	100	100	100
Scenedesmus ecornis	78	83	100
Scenedesmus denticulatus	57	72	100
Desmidiales			
Cosmarium cf *regnesii*	21	100	62
Staurastrum pseudopelagicum	100	72	30
Chrysophyceae			
cf *Monochrysis* sp	100	51	79
Pseudokephyrion planctonicum	100	68	54
Bacillariophyceae			
Nitschia actinastroides	2	34	100
Synedra nana	2	35	100

was raised from 3 to 6 µg/l. Also in contrast to the development in Lake Ömmern the trophic indexes of these lakes were markedly higher in 1979—81 (Fig. 3) as a result of the increased precipitation and run off, which improved the phosphorus supply after the dry years 1975—76.

After liming in Lake Ömmern 1981 the con-

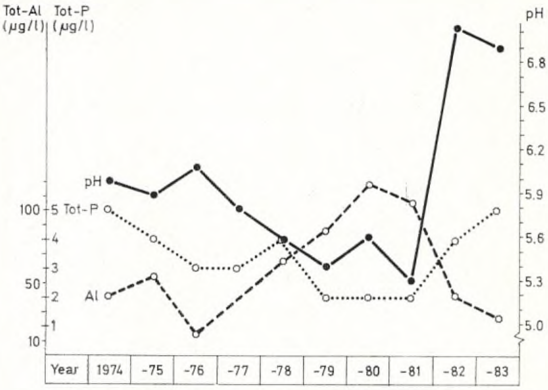


Fig. 2. Levels of pH, aluminium and phosphorus during the period 1974—83 in Lake Ömmern. (Average values July—August.)

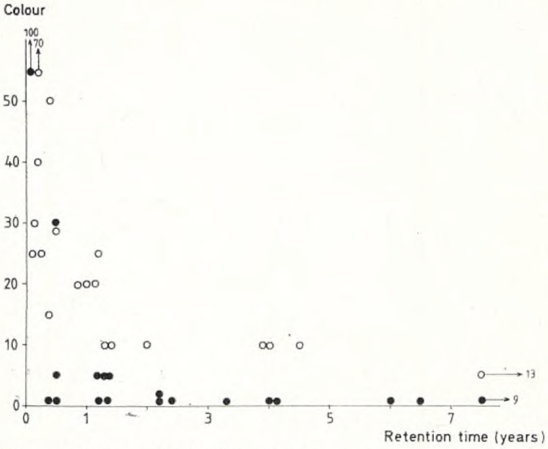


Fig. 4. The relationship between water colour, pH and water retention time. ●=pH < 5.5 ○=pH > 5.5.

centration of aluminium decreased to 20 µg/l and the concentration of total phosphorus was raised to the same level as before the acidification process started. The improved nutrient supply resulted in a change in the average number of species from 38 during the period 1978—81 to 57 in 1983. A great deal of these returning species were diatoms, which constituted the bulk of the phytoplankton biomass in 1983, as during the period 1973—76, when there was also a low level of aluminium.

All the strongly acid lakes with a water retention time longer than a year had in August 1976 an extremely clear, blue-green water (colour < 5), whereas lakes with pH-values above 5.5 had higher humus concentrations (Fig. 4). The average water colour of the west coast lakes was relatively high in 1971 and 1981 compared to 1976, when there was little run off. In the most acid lakes, however, the colour did not change (Fig. 5). Laboratory investigations indicated that the low level of humus compounds in the acid waters is not a result of low pH, but of high levels of aluminium. Both acid lake's aluminium and synthetic $Al_2(SO_4)_3$ caused a markedly raised precipitation of humus (Fig. 6).

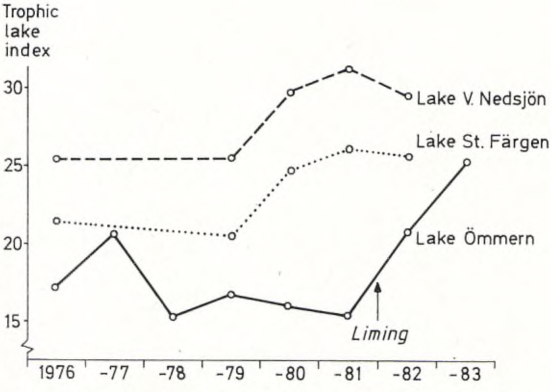


Fig. 3. Trophic lake index in Lake Ömmern and two adjacent lakes with pH around 6.5 during the period 1976—83. (Average values July—August.)

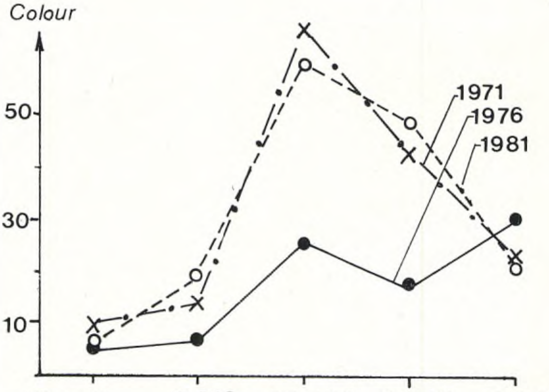


Fig. 5. Water colour in relation to pH in Swedish west coast lakes in August 1971, 1976 and 1981.

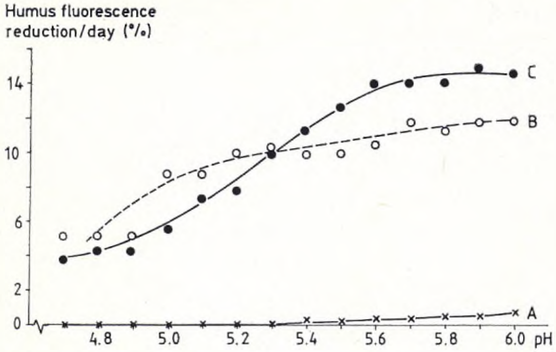


Fig. 6. Reduced humus fluorescence at different pH-values in lake waters with low (A=20µg/l) or high (C=300 µg/l) concentrations of aluminium, or synthetic water added with 300 µg Al/l (B).

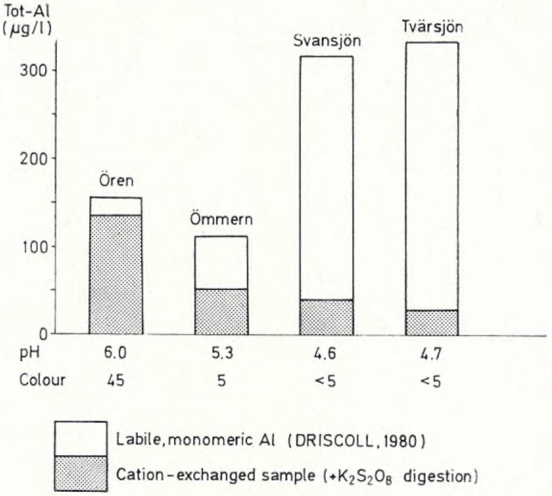


Fig. 8. The distribution of organically bound and labile aluminium respectively in relation to water colour and pH in four Swedish west coast lakes.

The distribution of phytoplankton species had a connection to water colour. Thus *e.g. Dinobryon borgei, D. bavaricum, D. divergens* and *D. suecicum* only occurred in acid lakes with a water colour of 30 or higher (Fig. 7).

The aluminium concentrations in the investigated lakes increase with the size of the drainage area, and in proportion to the run off. In the dry summer of 1976, the levels of aluminium in the strongly acid lakes generally were 100—500 µg/l. In some clear and acid (pH < 5.3) lakes, however, the levels were only about 50 µg/l. In these, 12 species of phytoplankton were registered (Table 2) which have not been observed in lakes with aluminium levels above 100 µg/l. One example of this is *Asterionella formosa* which constituted the bulk of the biomass in spring in Lake St. Härsjön at pH 4.8. Another 15 species were registered (Table 2) in humic water (colour $\geq$ 30), *e.g. Staurodesmus* spp.

Aluminium fractionation by cation exchange technique (Driscoll 1980) showed that about 90 % of the total aluminium in the clear waters of two acid lakes was in the form of labile monometric aluminium (Fig. 8), while the corresponding percentage level in Lake Ömmern with colour 5 was 50 %. The

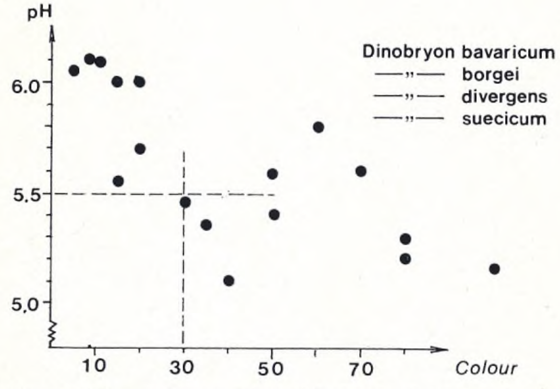


Fig. 7. The occurrence of *Dinobryon bavaricum, D. borgei, D. divergens* and *D. suecicum* in relation to water colour and pH-value in August 1976.

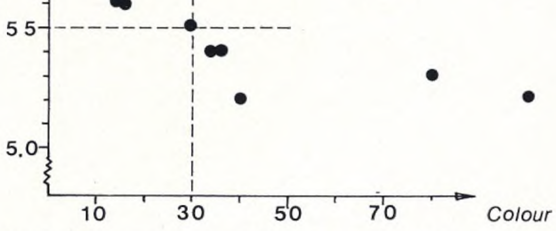


Fig. 9. The occurrence of *Staurodesmus* spp in relation to water colour and pH-value in August 1976.

Table 4. *The growth reduction of 19 species of phytoplankton exposed to aluminium ($Al_2(SO_4)_3$) in lake water of pH 5.5.*

Al (µg/l)	100	200	400
Chlorophyceae			
Crucigenia tetrapedia	—	100	100
Dictyosphaerium elegans	0	100	100
Eudorina elegans	0	0	0
Monoraphidium dybowskii	0	0	0
Monoraphidium griffithii	0	0	100
Scenedesmus ecornis	—	37	62
Scenedesmus denticulatus	27	73	91
Desmidiales			
Cosmarium regnesii	32	100	100
Staurastrum pseudopelagicum	—	100	100
Staurodesmus sellatus	—	100	100
Chrysophyceae			
Chrysococcus sp	0	0	0
Monochrysis sp	0	0	0
Ochromonas sp	—	100	—
Ochromonas sp	—	0	—
Pseudokephyrion planctonicum	0	0	0
Diatomeae			
Asterionella formosa	—	100	100
Nitschia actinastroides	47	100	100
Rhizosolenia longiseta	—	100	100
Synedra nana	47	100	100

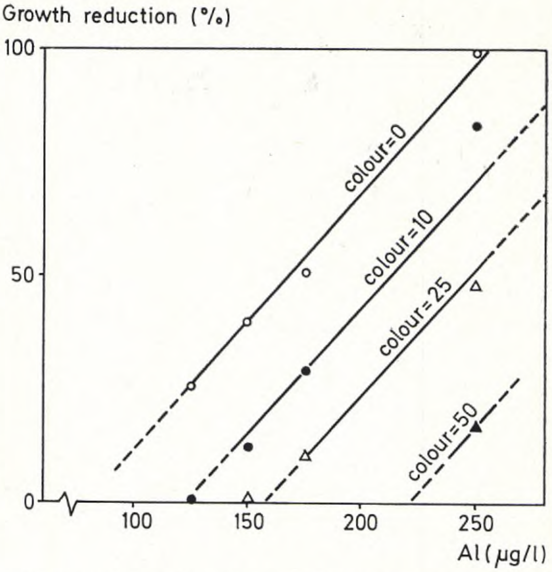


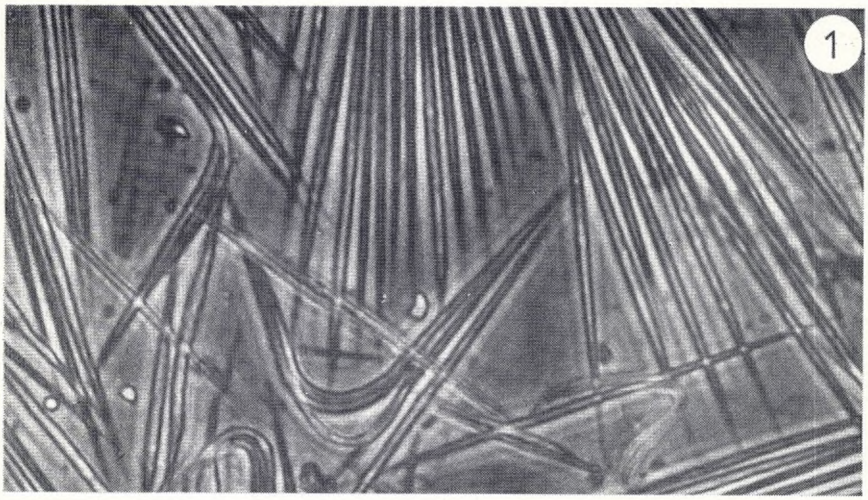
Fig. 10. *Monoraphidium griffithii* (Chlorophyceae). Percentage growth reduction in clear lake water of pH 5.5 and varied additions of aluminium and humus.

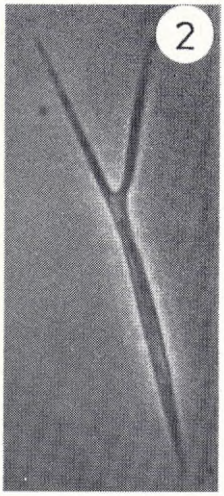
level of labile aluminium in Ömmern then was about 50 µg/l during the period 1979—81, when the pH-value (5.3—5.6) was in the range of maximal specific aluminium toxicity. During this period all desmids were eliminated. The most frequent of these species before the acidification were *Staurodesmus* spp and *Staurastrum anatinum.* At the inventory of lakes in 1976, however, it was apparent that the genus *Staurodesmus* occurred in the pH range 5.3—5.6 (Fig. 9). These lakes were, however, humic ones (colour ≥ 30). Also *Staurastrum* appeared to be equally acid tolerant, and was also registered in clear lakes, but only in those with aluminium levels below 100 µg/l.

Biotests with 19 algal species were carried out to investigate whether aluminium toxicity is a potential hazard in lakes. One of the biotests showed that 13 of them, especially desmids and diatoms, were markedly sensitive to aluminium (Table 4), since their growth was completely inhibited at 200 µg Al/l. The growth of *Scenedesmus* was reduced already at 100 µg Al/l but was still active at 400 µg/l. The concentration difference between growth reduction and inhibition was on the other hand, very small concerning desmids and diatoms (100—150 µg/l).

Monoraphidium dybowskii, occurring in aluminium-rich lakes, was unaffected by 400 µg/l as most crysophyceans. *Monoraphidium dybowskii* was optimally growing in water from Lake Tvärsjön with the addition of phosphorus. Thus it should be a good nutrient solution also to other species. In spite of that the growth of *Monoraphidium griffithii* was completely inhibited, which was also the case in water from Lake Härsevatten (pH 4.4). These two waters are rich in Al, 270 and 660 µg/l respectively. When diluting the waters with lake water poor in aluminium (10 µg/l) the growth started, but it was still reduced at 100 µg Al/l by a share of 15 % of water from Lake Härsevatten. The growth reduction of *Monoraphidium griffithii* in such dilutions and corresponding additions of synthetic $Al_2(SO_4)_3$ to Al-poor lake water, was about the same.

Monoraphidium griffithii occurred at about 100 µg tot-Al/l in Lake Ömmern 1981, which is in accordance with the Al-tolerance found in





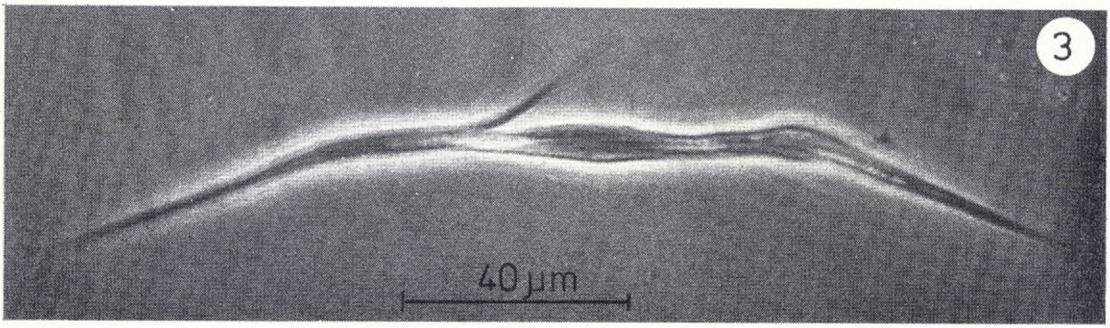


Plate 1. *Monoraphidium griffithii.* Normally dividing cells in lake water of pH 5.5 without aluminium. (Photo G. Rosén.)
Plate 2. *Monoraphidium griffithii* exposed to 150 µg Al/l ($Al_2(SO_4)_3$) in lake water of pH 5.5 (Photo G. Rosén.)
Plate 3. *Monoraphidium griffithii* exposed to 200 µg Al/l ($Al_2(SO_4)_3$) in lake water of pH 5.5 (Photo G. Rosén.)

laboratory tests. In extremely clear acid lake water, without humus, the growth was, however, reduced already at 80 µg Al/l. At addition of humus making a water colour of about 5, the growth was again normal. Fig. 10 shows that the concentration of humus has a decisive influence on the toxicity of aluminium.

The effect of aluminium was not manifested only as a growth reduction. The cells of *Monoraphidium griffithii* which are normally small and needle shaped (Plate 1) were enlarged already at aluminium concentrations below those causing reduced growth rate. At a higher Al-concentration other reproductionary disturbances were more frequent. At about 150 µg/l, cells with splitted tips occurred (Plate 2). The most characteristic effect, noticed when the growth was nearly inhibited, at about 200 µg Al/l, was the contortion of cells (Plate 3).

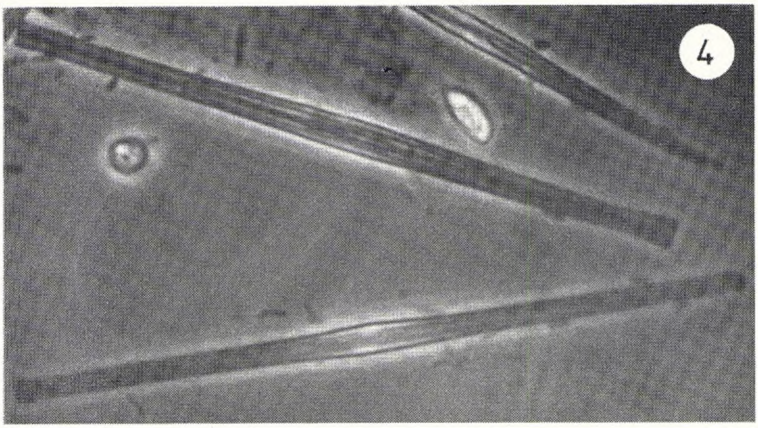
The cell structure of *Synedra cf nana* (Plate 4) was desorganized and the cell components formed droplets (Plate 5 A) already at 100 µg Al/l. Bubble formation was also noticed on the silicon shells (Plate 5 B).

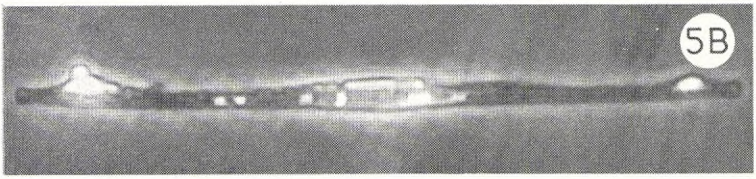
There was no distinctive difference between synthetic solutions of $Al_2(So_4)_3$ and the natural aluminium of Lake Härsevatten, in their inhibitory effects.

Zooplankton

Lake investigations

In the most acid (pH < 5.3) lakes there were generally few zooplankton species in August 1976. *Daphnia* spp and *Heterocope appendiculata* were





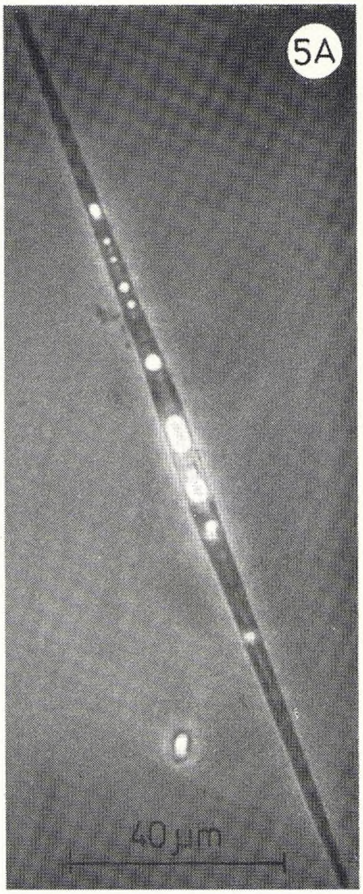


Plate 4. *Synedra* cf *nana*. Normal cells in lake water of pH 5.5 without aluminium. (Photo G. ROSÉN.)

Plate 5. *Synedra* cf *nana* exposed to 100 µg Al/l in lake water of pH 5.5. A). Lake water added with $Al_2(SO_4)_3$. B). Aluminium from acid lake. (Photo G. ROSÉN.)

lacking at pH-values below 6 and 5.75 respectively. At pH-values below 4.75 there were no registrations of *Conochilus spp*, *Trichocerca spp*, *Bythotrephes longimanus*, *Diaphanosoma brachyurum* and *Leptodora kindtii*. In 1979—81, however, *Daphnia cristata* and *D. hyalina* occurred frequently in Lake Ömmern at pH 5.3—5.6. According to all observations in acid lakes 1976—81, these appeared to be almost as rich in species as waters of pH 6—7.5 (Fig. 11).

With the exception of the extremely oligotrophic and strongly acid lakes, most zooplankton species occurred independent of the trophic state. Species of *Daphnia* and *Trichocerca*, however, were most abundant in lakes with relatively high trophic index values (21—40), while *Bythotrephes* had the opposite distribution (index 11—20) (Fig. 12). Excluding the most acid lakes, there was no correlation between the trophic lake index and the number of zooplankton species (r=0.08) (Fig. 13). There was, however, a connection between the species composition of zoo- and phytoplankton in acid lakes. In the more nutritious lakes *Conochilus* and *Keratella* occurred together with small chlorophyceans and chrysomonades, while in the most oligotrophic acid waters *Asplanchna priodonta*, *Bosmina coregoni* and *Polyarthra remata* were abundant when the phytoplankton biomass was dominated by dinophyceans (Table 5).

Ascomorpha, *Ceriodapnia* and *Daphnia* were most abundant in humic lakes, while *Bythotrephes* on the other hand required less humic waters (colour < 15). *Asplanchna* was curiously enough common partly in extremely clear acid lakes and partly in neutral but humic waters.

In lakes with aluminium concentrations exceeding about 180 µg/l markedly fewer species were registered (Fig. 14). Around 10 out of ca 30 totally registred zooplankton species were lacking, *e.g. Daphnia* spp, already at levels above 50 µg/l.

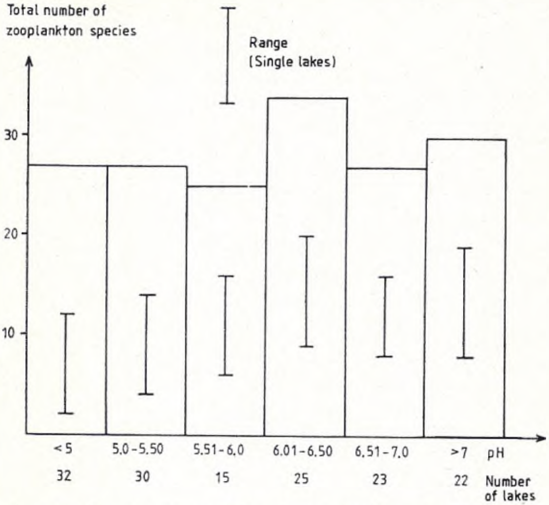


Fig. 11. The total number of zooplankton species registered at different pH-intervals.

Biotests

At the addition of 300 µg Al/l into a lake water of pH 5.5, all individuals of *Acroperus harpae* were dead after five weeks of exposure. In the control vessels the originally five animals had reproduced and at the end of the experiment there were 10, 12 and 25 individuals, respectively.

The percentage of surviving individuals of

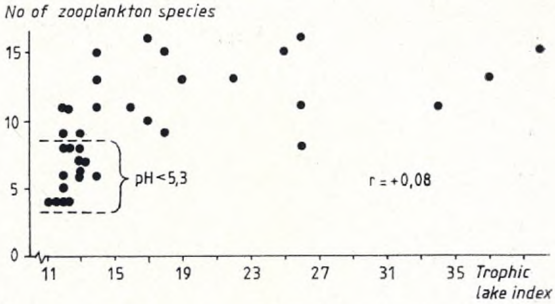


Fig. 13. Number of zooplankton species in relation to trophic lake index.

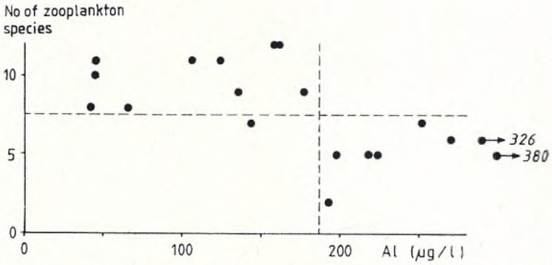


Fig. 14. The number of registered zooplankton species in acid lakes, in relation to the concentration of aluminium, in August 1976.

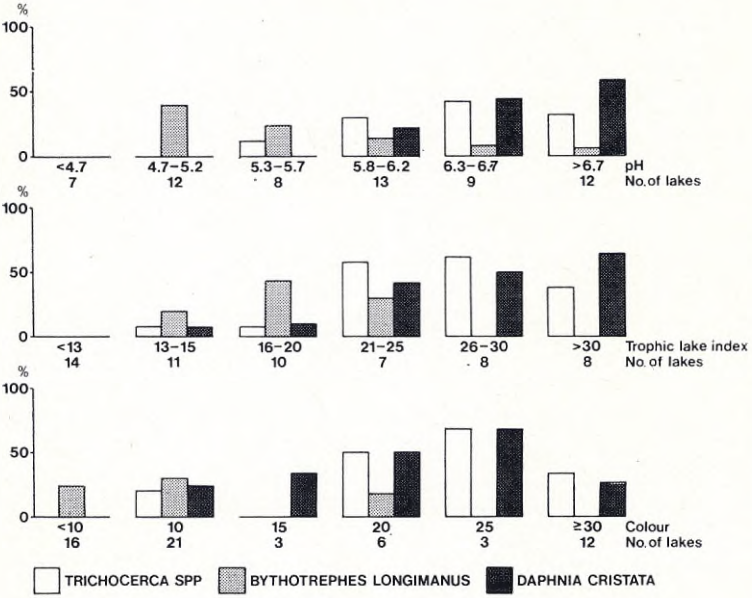


Fig. 12. Distribution of *Trichocerca* spp, *Bythotrephes longimanus* and *Daphnia cristata* in 60 lakes in the Swedish west coast area, in relation to pH, trophic lake index and water colour. (The abundance of the species has not been considered.)

Table 5. *Predominant phyto- and zooplankton species in acid lakes of different trophic levels in August 1976.*

Trophic lake index 11		Trophic lake index 15—16	
Dominating phytoplankton	Dominating zooplankton	Dominating phytoplankton	Dominating zooplankton
Lake Tvärsjön		Lake Rishagerödvatten	
Peridinium	*Asplanchna priodonta*	Dinobryon	*Keratella cochlearis*
Gymnodinium	*Polyarthra remata*	Ochromonas	*Ceriodaphnia quadrangula*
	Bosmina coregoni	Oocystis	*Diaphanosoma brachyurum*
Lake Stockasjön		Lake Buvattnet	
Peridinium	*Asplanchna priodonta*	Sphaerocystis	*Conochilus hippocrepis*
Gymnodinium	*Polyarthra remata*	Botryococcus	*Holopedium gibberum*
	Bosmina coregoni	Chrysophyceae spp	*Eudiaptomus gracilis*
Lake S. Boksjön		Lake Skällingesjön	
Peridinium	*Polyarthra remata*	Chromulina	*Conochilus hippocrepis*
Gymnodinium	*Bosmina coregoni*	Peridinium	*Keratella cochlearis*
		Oocystis	*Holopedium gibberum*
Lake St. Tresticklan			*Eudiaptomus graciloides*
Peridinium	*Asplanchna priodonta*		
Gymnodinium	*Polyarthra remata*	Lake Storsjön	
	Bosmina coregoni	Oocystis	*Conochilus hippocrepis*
		Monoraphidium	*Holopedium gibberum*
		Ochromonas	*Eudiaptomus gracilis*

Daphnia magna exposed to 300 µg Al/l during 48 hours was markedly reduced compared to the Al-free controls. At the addition of 1000 µg/l all daphnids were dead after 6 hours of exposure (Fig. 15).

IV. DISCUSSION

Phytoplankton

In the most acidified and oligotrophic lakes the phytoplankton is exclusively represented by a low number of slowly growing, motile flagellates, which by means of vertical migration probably are able to facilitate their nutrient supply. The scarcity of species in these waters is to a certain extent attributed to the acidity as such, since 18 phytoplankton species apparently are eliminated already at about pH 6 (Table 1), and probably some more of them at 5.7—5.0 (Table 3) as an effect of the strongly raised concentration of hydrogen ions. A considerably more important mechanism in this connection is, however, the aluminium induced precipitation of phosphorus (ALMER *et al.*

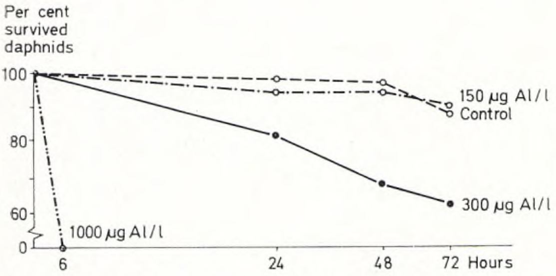


Fig. 15. *Daphnia magna* exposed to the aluminium concentrations 150, 300 and 1000 µg/l, during 72 hours in lake water of pH 5.5.

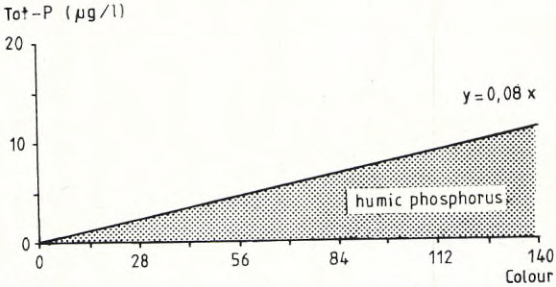


Fig. 16. The minimum concentration of total phosphorus, in relation to the water colour of 1250 Swedish lakes. (Unpublished data from W. DIETRICHSON and the Swedish lake investigation 1972.)

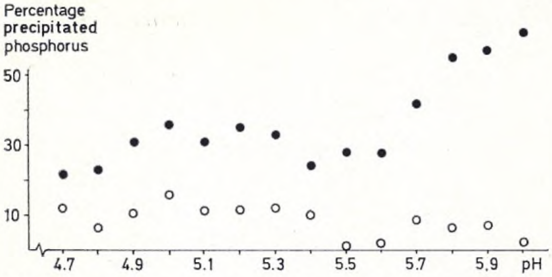


Fig. 17. Percentage of precipitated phosphorus (total values) of the original 100 µg/l, after three days at different pH-values in water from Lake Tvärsjön (colour=0) (●) and the same water of colour=10 achieved by addition of humus (○).

1978), which supply is the primary limiting factor of the algal growth in the studied lakes (Hörnström and Ekström 1983). Most phytoplankton species have a varying minimum requirement of nutrients (Hörnström, 1981; Rosén, 1981) and thus the supply of phosphorus, is an important factor which decides the species richness in the studied lakes. An example of this, is the development in Lake Ömmern (Fig. 2), where 50 % more species were registered as a result of the increased level of phosphorus from 2 to 5 µg/l (Hörnström and Ekström 1985).

It is a well-known fact that several phytoplankton species require a minimum concentration of humus (Pringsheim 1954), which seems to have a nutritional importance by complexing and facilitating the availability of nutrients. In the most acid lakes the level of humus is generally below these minimum requirements and this is probably also a reason for the reduced supply of phosphorus. An indication of this is namely the results of an investigation of 1250 Swedish lakes in 1972 (unpubl. data) from which it was obvious that a certain water colour was equivalent to a minimum level of total phosphorus (Fig. 16). That humic compounds have capacity to reduce the precipitation rate of phosphorus is also indicated from our laboratory experiments, where just a little content of humus (colour=10), had a marked effect (Fig. 17). Thus the presence of humus is of utmost nutritional importance to phytoplankton.

Oligotrophic and acid tolerant plankton species suddenly disappear when the concentration of aluminium is raised. In Lake Ömmern all desmids were eliminated when the Al-level had reached 100 µg/l and in other lakes with higher levels still more oligotrophic species were lacking in spite of their tolerance of acidity and oligotrophy. Desmids, however, occurred in lakes with low levels (ca 50 µg/l) of Al and in lakes where humus compounds could bind the Al-ions (Fig. 9). Diatoms and several species of chlorophyceae were also registered in these lakes (Table 2) only, all which indicates that aluminium is hazardous to phytoplankton. The registered connection between the Al-level and the occurrence of desmids and other species is supported by the results from our laboratory experiments, which showed that desmids are sensitive to aluminium. Already a concentration of 100 µg/l, markedly reduced the growth rate of two desmids (Table 4). This result is, however, in contrast to previous studies (Burrows Dickinson 1977) where desmids were markedly more tolerant than other species. These desmids were, however, benthic ones, which have probably different physiological characters compared to real plankton species. Several biotests in this investigation have shown that the range of maximal specific aluminium toxicity is found in the pH interval 5—6, while the toxicity is decreased at higher pH-values. Previous biotest investigations (Burrows Dickinson 1977; Claesson and Törnqvist, pers. comm.) have been performed at pH 6—8, and this should be the main reason why the registered critical Al-levels were markedly higher. It is, however, also notable that the sensitivity among species, also closely related ones, is varying.

Common features of phytoplankton exposed to this metal are growth reduction and enlarged cell volume, verified also in other investigations (Claesson and Törnqvist pers. comm.). Most remarkable is, however, the complete destruction of cell components of diatoms, the physiological mechanisms of which are still unknown.

13 out of 19 tested species showed a reduction or inhibition of their growth at Al-concentrations of 100—200 µg/l. Assuming that the tested species are representative of the 130 phytoplankton forms registered in the studied lakes, this means that 68 % of the species are eliminated by increased aluminium toxicity. The percentage share is, however, probably not so high, since the distribution of several of these species, *e.g. Dictyosphaerium* is primarily limited by a low supply of nutrients, or

low pH. The acid induced elimination of phytoplankton species thus is an interplay between high acidity, nutrient depletion and aluminium toxicity, so it is difficult to quantify the separate effects. The increased concentration of aluminium, however, is mainly responsible for the disappearance of those ca 100 species which are lacking in the ultraoligotrophic acid lakes.

Zooplankton

A survey of different investigations (HOBAEK and RADDUM 1980) indicates that most zooplankton species may occur in acid lake waters of pH 4.5—5.3. In the 1976 investigation *Bythotrephes longimanus* and some other species did not occur at pH-values below 4.75. On the other hand *Bythotrephes* had the largest abundance in lakes with pH 4.75—5.3, which may indicate that factors other than the acidity are responsible for the elimination of this and other species in strongly acidified waters. Most striking was the finding of daphnids at pH > 6 only. *Daphnia*, however, have been recorded at lower pH-values in other investigations (HOBAEK and RADDUM 1980) and in Lake Ömmern 1981. These observations at high acidity were generally made in more or less humic lakes, where the supply of food — detritus and bacteria — is relatively large compared with that in extremely clear lakes (JOHANSSON 1983). The scarcity of species in the west coast lakes in 1976 was, consequently not caused by high acidity, but is probably to a great extent a result of the aluminium induced oligotrophication process. The only zooplankton species which could be characterized as sensitive to low pH-values is *Heterocope appendiculata*, which was registered only at pH > 5.75.

The humus-limited occurrence of several species in highly acid waters and the fact that toxic Al-ions are effectively bound to the humic compounds (Fig. 8), suggest that high levels of aluminium cause the elimination of species in clear lake waters. This is also indicated by the results from biotests with *Daphnia* and *Acroperus*, which showed the same critical Al-level (150—300 μg/l) as the 1976 lake investigation.

The sensitivity of *Daphnia magna* to aluminium has previously been investigated (BURROWS DICKINSON 1977). The toxic effects were registered at higher levels of aluminium than in this investigation. This probably was a result of the high pH-value (6.5—7.5), which exceeds the pH-range of maximal specific aluminium toxicity found at pH 5—6.

In conclusion, the high supply of aluminium to the acid lakes is, through oligotrophication and toxicity, alone responsible for the sparse zooplankton fauna.

V. REFERENCES

ALMER, B., W. DICKSON, C. EKSTRÖM and E. HÖRNSTRÖM. 1978. Sulfur pollution and the aquatic ecosystem. p. 271—311. *In* Sulfur in the environment. Part II. Ecological impact. Ed.: J. O. Nriagu. John Wiley & Sons, New York.

BURROWS DICKINSON, W. 1977. Aquatic Aluminium: Chemistry, toxicology, and environmental prevalence. CRC Critical Reviews in Environmental Control: 167—216.

DRISCOLL, C. T. 1980. Chemical characterization of some dilute acidified lakes and streams in the Adirondack region of New York State. Thesis, Cornell Univ. 296 p.

HOBAEK, A. and G. G. RADDUM. 1980. Zooplankton communities in acidified lakes in south Norway. SNFS-project IR 75/80. Oslo—Ås. 132 p.

HÖRNSTRÖM, E. 1981. Trophic characterization of lakes by means of qualitative phytoplankton analysis. Limnologica 13: 249—261. (In English with German summary.)

— and C. EKSTRÖM. 1983. pH-, närings- och aluminiumeffekter på plankton i västkustsjöar. Nat. Swedish Environm. Protec. Bd SNV PM 1704. 124 p. (In Swedish with English summary.)

— and C. EKSTRÖM. 1985. Acidification and liming effects on phyto- and zooplankton in some Swedish west coast lakes. Nat. Swedish Environm. Protec. Bd. SNV PM. 1864.

JOHANSSON, J.-Å. 1983. Seasonal development of bacterioplankton in two forest lakes in central Sweden. Hydrobiologia 101: 71—88.

NRCC. 1981. Acidification in the Canadian environment. National Research Council of Canada, Associate Committee on Scientific Criteria for Environmental Quality. NRCC 18475. 369 p.

PRINGSHEIM, E. G. 1954. Algenreinkulturen ihre Herstellung und Erhaltung. Veb Gustav Fisher Verlag. Jena. 109 p. (In German.)

ROSÉN, G. 1981. Phytoplankton ecology — results from the "Thousand Lakes' investigation". Rep. Nat. Swedish Environm. Protec. Bd. 119 p. (In Swedish with English summary.)

RÖNDELL, B. 1982. List SO keywords and codes for water research. Nat. Swedish Environm. Protec. Bd SNV PM 1492. 277 p. (In Swedish with English summary.)

WILLÉN, E. 1974. Methods of phytoplankton investigations. Nat. Swedish Environm. Protec. Bd. PM 525, NLU Rep. 76. 45 p. (In Swedish with English summary.)

Reclaiming Acid High Mountain Lakes by Liming: A Progress Report

TOROLF LINDSTRÖM,[1] WILLIAM DICKSON [2] and GÖTE ANDERSSON [3]

[1] Institute of Freshwater Research, S-170 11 Drottningholm, Sweden.
[2] National Swedish Protection Board, P. O. Box 1302, S-171 25 Solna, Sweden.
[3] Domänverket (The Swedish Forest Service), S-791 81 Falun, Sweden.

ABSTRACT

Ranking different fish species according to a pH-tolerance list has proved to be very useful as a framework for studying the acidification process. Experimental studies have, however, shown that the aluminium concentration has an important effect on fish survival.

The lakes in the Fulufjäll high mountain range are acidified and a liming programme has been applied since the beginning of the 1970s. The fluctuations in the year classes of char in the acid Lake Stora Rösjön have been compared to the concentrations of dissolved plus suspended aluminium. Good year classes occur when the aluminium concentration is not rising above 70 µg/litre in the second half of the first year of life of the fish. This is a very low concentration, but most of the aluminium should occur in unchelated form. A poor length-weight relation resulted when the aluminium concentration was above 50 µg/litre during most of the early life of the young char.

New char populations were established after liming in two cases, when the aluminium concentration was below 70 µg/litre in the second half of their first year of life. Char had become extinct in the Särnamannasjöarna lakes. The upper lake runs through a short and fairly slow brook down to the lower lake, which is in most respects very similar to the upper lake. When the downstream lake was limed and stocked with char in 1975, a population was established. The aluminium concentration rarely exceeded 50 µg/litre, but when it did so in 1981 and 1982, the condition of the young char was very poor. There were no competing fish species in the lake. The char did not migrate to the untreated upper lake.

Adult char had been living in Lake Stora Harrsjön since 1964, but the population did not reproduce. After liming in 1976, the aluminium concentration did not exceed 70 µg/litre in the second halves of 1977 and 1978, but char reproduction was not successful until 1980. It is probable that the native trout in this lake delayed char reproduction for some years by competitive interaction. The year class of 1980 had a very good lenght-weight relation, although they had been exposed to aluminium values of about 50—70 µg during the whole of their life. The char populations are thus somewhat different in this respect in the three lakes.

Different fish populations utilize the available resources very differently in acidified lakes, with regard to population density, growth and migration. Therefore, the growth of fish populations in different acid lakes may vary greatly.

I. INTRODUCTION

The sedimentary Jothnian rock and the quarternary deposits in the Fulufjäll mountain range in western Sweden contain very little lime. Acid precipitation would have a serious effect on the water quality of the lakes and running water. This latent risk was recognized at an early stage and a liming programme was gradually started in 1972. The programme was urgently needed as the fish populations were on the verge of extinction. The Arctic char populations of the Särnamannasjöarna lakes and the grayling populations of the Harrsjöarna lakes were actually extinct at the beginning of the 1970s (LINDSTRÖM and ANDERSSON 1981). It is also probable that the trout populations of the Tangsjöarna lakes became extinct later on, although the material is less conclusive.

In this "borderland" the survival or extinction of a population of fish is not only controlled by the water quality but also by developments in the population ecology of the fish (*op. cit.*). The reclaiming of a lake by liming is not only attained by raising the pH over a level that has been described as a limit for the species.

Experimental studies have shown that the aluminium concentration has an important effect on fish survival (Schofield 1977, Dickson 1978 a, b, Leivestad *et al.* 1980). The aim of the present study has been to specify the aluminium concentrations that result in good or poor year classes of char and in good or poor condition in young char and the concentrations that influence the establishment of new populations.

This study is based on observations made during the past ten years, and constitutes a basis for further studies. The importance of population ecology in all these processes will be studied.

II. MATERIAL AND METHODS

The study concerns Lake Nedre Särnamannasjön, Lake Stora Rösjön and Lake Stora Harrsjön (Fig. 1). The present investigation is a continuation of a previous investigation, carried out in the same district (Lindström and Andersson 1981). The water quality and the liming measures are indicated in Figs. 2—4. The liming methods have changed somewhat during the course of the studies. Only the resulting water quality is reported. The water analyses have been performed according to the Swedish Standard Methods' (1981) recommendations. The methods used for the analysis of the fish stocks are the same as reported in Tables 3—6 of the previous publication.

The morphology and ecology of the two types of char found in the district have been described in detail in the previous publication. Nyman *et al.* (1981) have established their systematic position as two sibling species, normal char *Salvelinus salvelinus* and F-char *Salvelinus alpinus*.

The number of char caught in Lake Stora Rösjön in two gill nets of mesh size 16.7 mm knot-to-knot, has been used to provide an estimate of the strength of the different year classes (Table 1). These gill nets were set in a standardized way, at the end of August—beginning of September, after the spawning of the dwarfed F-char. A number of the char were aged, and the results were used to estimate the age of the total catch in the two gill nets. Summing over all catch years gives an estimate of the strength of a year class.

The Fulton coefficient is expected to vary with the total length of the fish, state of the fish (immature, spawning etc.), season and the gear used for the catch, but this does not always occur. Four-summer-old char from Lake Stora Rösjön, caught in four different mesh sizes in August 1979 were compared. The mean length increased from 27 to 32 cm with increasing mesh size and the Fulton coefficient increased from 9.34 to 9.84. However, no such tendency was evident for the four-summer-old char in September 1981.

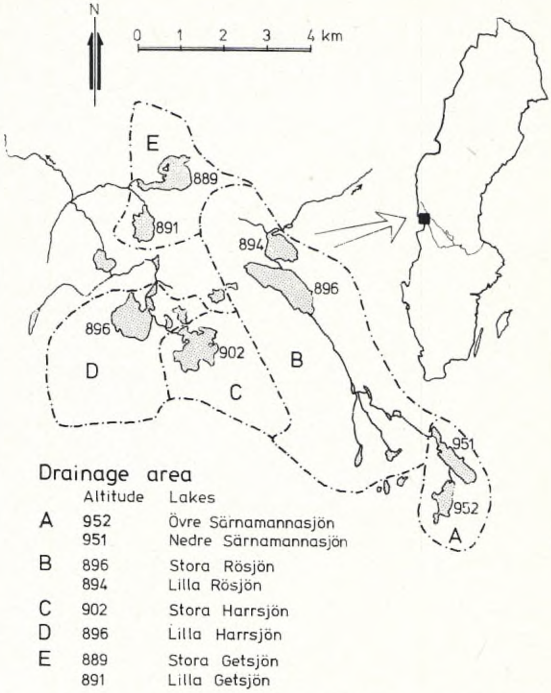


Fig. 1. Map of the lakes in the Fulufjäll mountain range and their drainage areas.

The condition of three-summer-old immature normal char, caught in gill nets of mesh size 16.7 mm in Lake Stora Rösjön in August/September, 1981 could be compared to that of a corresponding group from the year class of 1977, caught in the same lake in 1979. The very poor condition of the char in Lake Nedre Särnamannasjön from the year class of 1979, caught in 1981, cannot be compared in a corresponding way, as the fish introduced in the empty lake in 1975 and 1976 had grown extremely well, and suitable material is lacking from the following years.

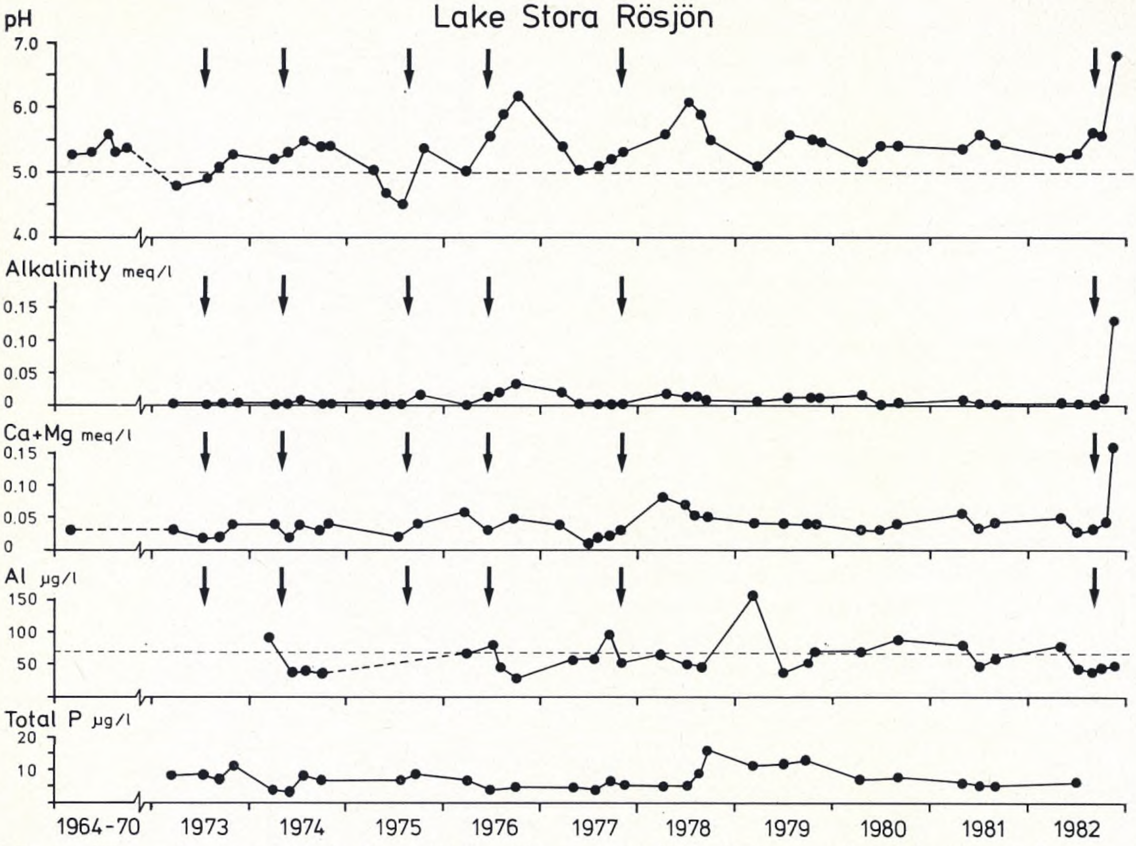


Fig. 2. Water chemistry in Lake Stora Rösjön. The arrows indicate liming occasions. The lake receives water from Lake Nedre Särnamannasjön, cf. Figs. 1 and 3.

Table 1. *The strength of year classes of char in Lake Stora Rösjön. Samples of normal char (N) and F-char (F) from gill nets with mesh size 16.7 mm were aged, and the results were used to estimate the age of the total catch in two such gill nets. The two nets were set in a standardized manner.*

Date	Number of char in the aged sample	Number of char in the catch	Year class:										
			1971	1972	1973	1974	1975	1976	1977	1978	1979	1980	1981
Aug. 30, 1977	7 N	7 N	—	—	2	—	2	3	—	—	—	—	—
	16 F	43 F	8	3	19	3	5	5	—	—	—	—	—
Aug. 29, 31, 1978	6 N	7 N	—	1	1	2	—	3	—	—	—	—	—
	12 F	68 F	—	—	—	—	23	45	—	—	—	—	—
Aug. 28, 30, 1979	22 N	22 N	—	—	—	—	1	4	17	—	—	—	—
	23 F	33 F	—	—	—	—	1	30	1	1	—	—	—
Sep. 4, 5, 1980	7 N	8 N	—	—	—	—	—	—	1	7	—	—	—
	26 F	59 F	—	—	—	—	2	28	3	24	2	—	—
Sep. 1, 3, 1981	25 N	25 N	—	—	—	—	—	—	—	5	20	—	—
	28 F	45 F	—	—	—	—	—	5	5	19	16	—	—
Sep. 10, 1982	15 N	15 N	—	—	—	—	—	—	—	1	6	7	1
	27 F	107 F	—	—	—	—	—	—	4	20	79	4	—

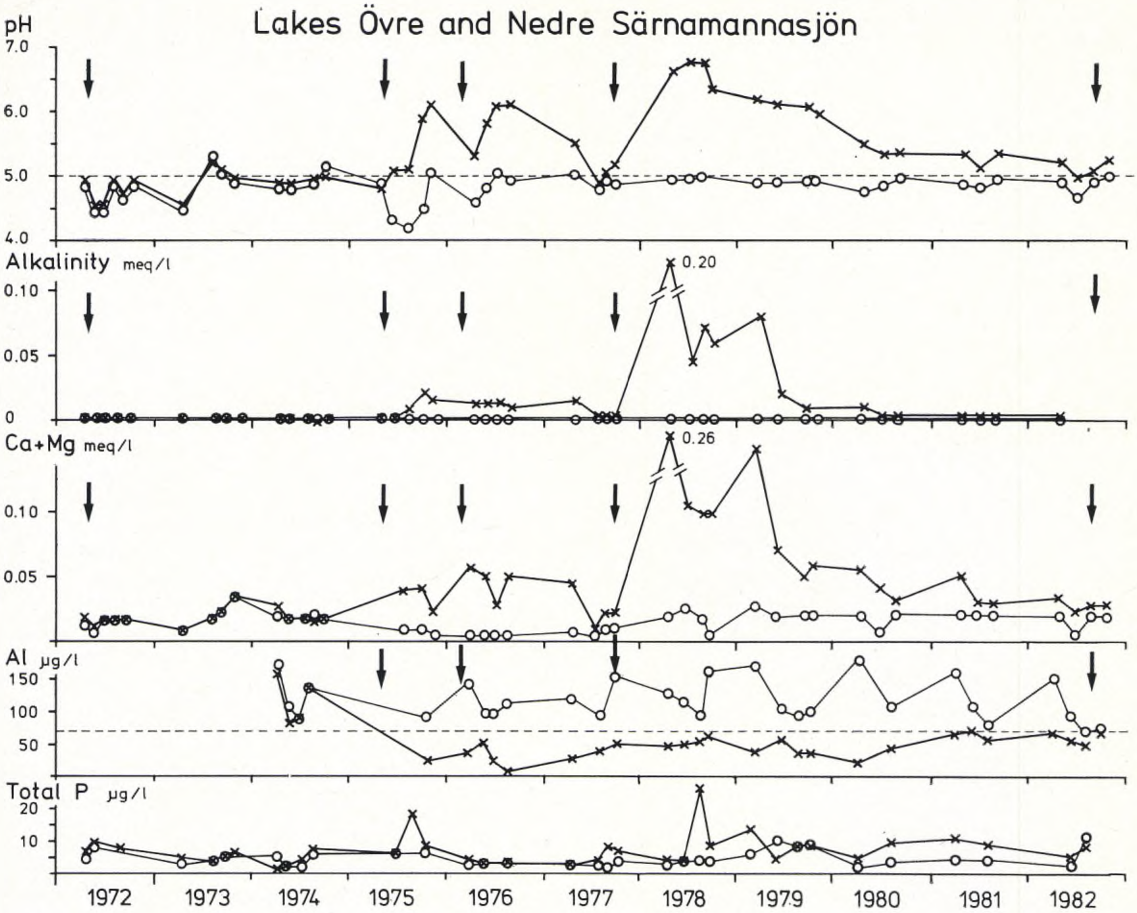


Fig. 3. Water chemistry in Lake Nedre Särnamannasjön (crosses) and water chemistry in Lake Övre Särnamannasjön (dots). The arrows indicate liming occasions.

III. RESULTS

The liming of the inlet brook to Lake Nedre Särnamannasjön in 1972 had almost no effect on the lake, as can be seen by a comparison of the water quality with that of Lake Övre Särnamannasjön, the untreated uppermost lake in the system (Fig. 3). As a result of later limings, the pH, alkalinity, calcium plus magnesium content and total phosphorus content have increased in Lakes Stora Rösjön and Nedre Särnamannasjön, whereas suspended plus dissolved aluminium has decreased (Figs. 2 and 3). The water analyses from Lake Stora Harrsjön are incomplete prior to 1976. An increase in the pH was registered, cf. Table 2 in the previous publication (Lindström and Andersson 1981) and Fig. 4 in the present paper.

The procedure for estimating year-class strength in Lake Stora Rösjön, presented in the previous chapter, indicates that the year classes of 1976 and 1979 were the most important ones in the period studied (Table 1).

In Lake Nedre Särnamannasjön, attempts were made to establish a char population in 1975 and 1976 (*op. cit.*, 1981). The development of the introduced char population in this lake is here illustrated by the catch in gill nets of mesh size 27.3 mm knot-to-knot and finer (Table 2). No other fishing took place in the lake during this period, which makes a population estimate easier.

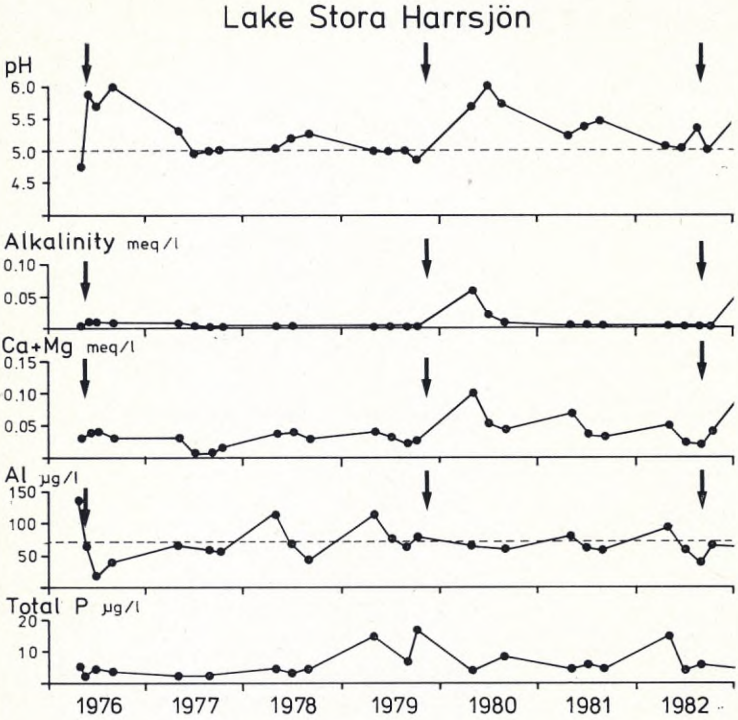


Fig. 4. Water chemistry in Lake Stora Harrsjön. The arrows indicate liming occasions.

The 1979 year class was the first important year class resulting from reproduction in the lake, forming 75 per cent of the catch in 1981 and 1982. Reproduction has, however, occurred during all years after 1976 according to information from age determination. In contrast to the situation in Lake Stora Rösjön, the normal char in Lake Nedre Särnamannasjön did not grow so well. These fish probably suffered from a high natural mortality in some years, as the catch in nets of mesh size 27.3 mm was slight in most years. However, a population of normal char has become established during the liming project. Single F-char were obtained in the catches in 1981—83.

Table 2. *The catch of normal char in gill nets in Lake Nedre Särnamannasjön. The catch in numbers per gill net effort. The mesh size is given in mm from knot-to-knot. The fishing time in hours varied between dates but not between nets set on the same date.*

Date	Number of char in nets of mesh size:				Number of nets of mesh size:			
	27.3	16.7	12.5	10.0	27.3	16.7	12.5	10.0
June 30, 1976	6.0	—	—	—	1	0	0	0
June 11. 1978	7.4	—	—	—	5	0	0	0
Sep. 21, 1978	3.7	—	—	5.0	3	0	0	2
June 10, 1979	5.0	4.0	2.0	—	1	1	1	0
Aug. 29, 1979	11.5	—	2.0	2.0	2	0	1	1
Sep. 2, 1981	2.0	32.0	9.3	—	1	2	3	0
Sep. 9, 1982	2.5	15.5	—	—	4	2	0	0
Sep. 3, 1983	11.5	30.0	—	—	4	2	0	0

Table 3. *Condition factor, (weight in g)×1000/(length in cm)^3, normal char from Lakes Nedre Särnamannasjön and Stora Rösjön. The char were caught in gill nets of mesh size 12.5—37.5 mm and by ice fishing.*

Lake	Date	Number of char	Mean length of char (cm)	Mean age of char	Mean condition factors from different types of gear						Notes
					mesh size, mm				various mesh size, mm	ice fishing	
					12.5	16.7	21.4	27.3	21.4—37.5		
Nedre Särnamannasjön	June 30, 1976	13	26.1	2+	—	—	8.59	—	—	—	
	Sep. 21, 1978	10	30.5	3+	—	—	—	7.75	—	—	spawning char
	Sep. 2, 1981	19	17.5	2+	—	6.91	—	—	—	—	
		8	14.9	2+	6.37	—	—	—	—	—	
	Sep. 9, 1982	3	16.2	2+	—	7.55	—	—	—	—	
		23	19.0	3+	—	6.48	—	—	—	—	
		8	21.8	3+	—	—	—	7.09	—	—	
Stora Rösjön	Aug. 28, 30, 1979	5	22.2	2+	—	—	8.84	—	—	—	
		17	19.0	2+	—	8.52	—	—	—	—	
		20	29.6	3+	—	—	—	—	9.63	—	
	April 5, 1982	42	15.5	—	—	—	—	—	—	7.70	all char
		13	18.9	—	—	—	—	—	—	7.89	those ≥ 16 cm selected
	Sep. 1, 3, 1981	5	21.8	2+	—	—	7.92	—	—	—	
		17	19.2	2+	—	7.61	—	—	—	—	
		20	29.4	3+	—	—	—	—	8.73	—	
	Sep. 10, 1982	6	18.7	2+	—	8.26	—	—	—	—	
		6	27.5	3+	—	8.96	—	—	—	—	

They have probably migrated upstream into the lake as the water quality became tolerable.

The length-weight relation indicated very poor condition in young normal char in Lake Stora Rösjön in 1981 and in Lake Nedre Särnamannasjön in 1981 and 1982 (Table 3).

The catch from fishing surveys in Lake Stora Harrsjön between 1964 and 1979 had consisted of more than 191 trout (*Salmo trutta*), 61 char and 2 grayling (*Thymallus thymallus*). Trout and grayling were the original inhabitants of the lake. As no young char were caught, although fine-meshed gill nets were included in the net set, the char were those stocked in the lake in 1964 or 1971. Residual eggs were often observed in female char caught in the spring. In 1982, 24 char and 80 trout were obtained in five gill nets. The char were mainly caught in nets of mesh size 27.3 mm, and all of them belonged to the 1980 year class. Their condition was good, as the length-weight coefficient was 9.59 for 22 fish with a mean total length of 29.6 cm. The liming programme carried out in 1976 and 1979 and the development of the water quality can be seen in Fig. 4.

The liming programme had included a tarn draining into Lake Stora Harrsjön, where no fish had ever been caught as far as we know. In 1983 trout had migrated up into this tarn and were caught by sport fishermen.

IV. DISCUSSION

The effect of aluminium concentrations on reproduction and condition

The discussion proceeds from the following points:

(1) The pH has not dropped below 4.5 in the lakes of the Fulufjäll mountain range, and a pH above 4.8 is not in itself a serious source of stress to the eggs and early stages of some salmonid fishes (Leivestad *et al.* 1980, Peterson *et al.* 1980, Runn 1982);

(2) An aluminium content of 70 µg/litre is a limiting concentration for sticklebacks. This level corresponds to ten days' survival (Erichsen Jones 1939, 1964). Lower levels and longer survival times were not studied;

(3) When the pH rises from 5 to 5.5 a substantial part of the dissolved aluminium is transformed into unionic but still reactive aluminium. It takes some time for the aluminium to precipitate and settle, and in the meantime it can still be poisonous to fish (Dickson 1983);

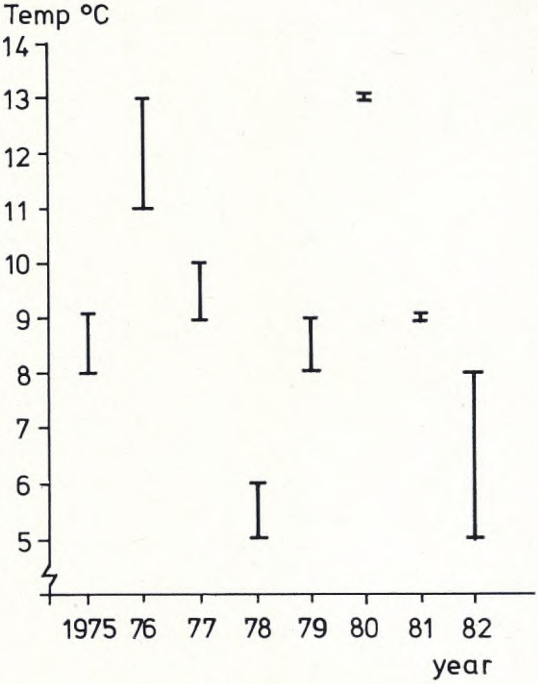


Fig. 5. The water temperature in the lakes on the Fulufjäll mountain range in August/September from 1976—81, on September 10 in 1975 and September 7—9 in 1982.

(4) Sensitivity to low pH decreases and sensitivity to the poisonous effect of aluminium increases with increasing age in the early stages of brook trout and white sucker (Baker and Schofield 1980 cf. Nelson 1980, Runn 1982).

In a lake, where the char live on the verge of extinction, a closer study of the water quality and reproduction in different years may indicate which factor is responsible for the failure or success of reproduction, as in Lake Stora Rösjön (Table 1).

The 1976 and the 1979 year classes are the two most important during the investigation period. Neither total phosphorus (Fig. 2) nor temperature shows a consistent positive relation to reproductive success. According to records of the air temperature at Särna, some 30 km away, 1976 was a rather normal year with a mean

August temperature one degree above normal, whereas in 1979 the mean air temperature was two degrees warmer in June but below normal in July and August. The water temperatures from the outflows of the Fulufjäll lakes agree well with this pattern: 1976 was warm but 1979 was colder than average in August/September from 1975—82 (Fig. 5). Calcium plus magnesium content (Fig. 2) might have affected food production and decreased the poisonous effect of aluminium, but the peaks in the graphs show no consistent relation to the years of reproductive success, 1976 and 1979.

Keeping strictly to the starting points of the discussion, the years 1976 and 1979 are the two years when the aluminium content did not exceed 70 µg/litre in the second half of the year from 1975—80 (the reproductive success in 1980 and 1981 has not yet been estimated).

Two comments must be added. The aluminium value discussed, 70 µg/litre, is very low, but aluminium should mainly occur in unchelated form as the levels of humic substances are very low. Thus, most of the aluminium is poisonous. Secondly, a field study without experiments cannot provide fully conclusive evidence. Further discussion of different fractions of dissolved or suspended aluminium will not be dealt with in this paper.

It may appear to be an uninteresting coincidence that this aluminium value, from a survival experiment with sticklebacks (ERICHSEN JONES 1939, 1964), should show any relation to the observations of intermittent reproduction in char in Lake Stora Rösjön. However, the importance of this aluminium level obtains some support from an analysis of the condition (length-weight relation) of the char.

If an aluminium value of 70 µg/litre limits reproductive success, a lower concentration, down to 50 µg/litre, could be suspected to affect metabolism and thus condition, as the gills are the organs affected in acidified water (LEIVESTAD *et al.* 1976, SCHOFIELD 1977, MUNIZ and LEIVESTAD 1980, LEIVESTAD *et al.* 1980). The length-weight relation as an estimate of condition has been criticized for giving high variability (ELLIOT 1976). In order to reduce the variability, FULTON coefficients are given for immature normal char, three summers old, taken from similar mesh sizes in Lake Stora Rösjön in August/September in 1979 and 1981 (Table 3). For nets of mesh size 16.7 mm the mean total char length was 19 cm and for mesh size 21.4 mm, the mean length was 22 cm in both years. A comparison of the water quality in Lake Stora Rösjön with the catches of the 1977 and 1979 year classes shows that the 1979 year class had spent almost two years in water with more than 50 µg aluminium/litre. The 1977 year class had experienced corresponding aluminium concentrations for a shorter duration, and their condition was better when they were caught.

The char populations in Lakes Övre and Nedre Särnamannasjön became extinct at the beginning of the 1970s, as the pH dropped below 5.0. According to present knowledge, the pH is not the only parameter of importance when describing the extinction of a populaion. Chemical factors which co-vary with pH, as well as diseases, competition from other species and fishing pressure can influence the extinction or survival of a population (LINDSTRÖM and ANDERSSON 1981). However, ranking fish species according to a pH-tolerance list has proved to be of very great value as a framework for studying the acidification process. There is little water quality data from the period when the char populations disappeared (*op. cit.*, Table 2), but the information in the present paper from Lake Övre Särnamannasjön (Fig. 3) gives some idea as to the situation in both lakes at the beginning of the 1970s. The water from Lake Övre Särnamannasjön passes through a brook of about 100 m length down to Lake Nedre Särnamannasjön, and their drainage areas are very similar.

The pH varies between 4.5 and 5 in Lake Övre Särnamannasjön and the aluminium level drops each year to about 100 µg/litre in the summer or autumn, but never falls below 70 µg.

Two attempts had been made to stock char in the lakes of the Fulufjäll mountain range (Lakes Getsjön and Stora Harrsjön), but self-reproducing populations were not established until a new introduction after the liming of Lake Nedre Särnamannasjön in 1975. The pH of this lake rose to 5.5—6, the aluminium level dropped below 50 µg/litre and the introduction was a success. The aluminium concentration stayed below 50 µg/litre until late in 1980. Spawning first occurred in

1976, although the bulk of the char introduced in 1975 spawned as four-summer-olds in 1977. Reproduction then took place in the lake each year, but the most important year class up to now is the 1979 year class, caught in 1981 and 1982 (Table 2). Of course, it takes some years to establish a new population. The effect of the water quality on the year classes of 1980 and 1981 can not be estimated as yet.

This latest introduction of char differs from the earlier ones in the sudden and important amelioration of the water quality and the lack of competing fish species.

However, in late 1980 and 1981 the year class of 1979 was exposed to an aluminium concentration above 50 μg/litre. The condition of the char caught in 1981 was poor, just as in Lake Stora Rösjön, but the exposure to high aluminium levels was of shorter duration in Lake Nedre Särnamannasjön. The aluminium concentration stayed above 50 μg/litre in 1982 also, and the condition of the char in Lake Nedre Särnamannasjön was poor in this year too (Table 3). The char caught in gill nets of mesh size 16.7 mm were now four summers old and had reached the same total length as three-summer-old char in Lake Stora Rösjön in 1981.

The introduction of char into Lake Stora Harrsjön sheds some more light on the establishment of a new char population. For the period 1964 to 1975 a complete analysis of water quality is lacking, but if the water quality is coarsely characterized by pH, Lake Stora Harrsjön resembled Lake Stora Rösjön in 1964—70. There was no indication of reproduction in the char transferred from Lake Stora Rösjön in 1964 and 1971 although reproduction occurred in the donor lake. After liming in 1976 the aluminium content of Lake Stora Harrsjön decreased to below 50 μg/litre, and did not exceed 70 μg/litre in the second halves of 1977 and 1978. However, it was not until 1982 that young char were caught in the gill nets, and these belonged to the year class of 1980 only. The earlier lack of reproduction in Lake Stora Harrsjön should partly have been the result of interaction by the trout population native to this lake. The char from the 1982 catch were in good condition. They had been exposed to aluminium levels of about 50—70 μg/litre during their whole life. There are differences between lakes in the development of the condition of young char. The char from the 1982 catch in Lake Stora Harrsjön were the first char born in the lake, and their good growth recalls that of the char stocked into Lake Nedre Särnamannasjön in 1975. The poisonous effect of the aluminium level is in some way reduced, perhaps through a surplus of food for young char.

If it is accepted that a trout population native to a lake offers resistance to the establishment of a new char population, then it can be assumed as a basis for further studies that a certain concentration of aluminium to some degree controls the reproduction of the char populations, and a lower aluminium concentration affects the condition of young char. The current aluminium concentrations are similar in the studied lakes. Deviations from this general pattern can be traced and should depend on differences between the lakes.

The strategy for utilizing the energy in acid lakes

During an episode of poor water quality, the reproduction of the char is damaged that year and the condition of immature char deteriorates. If such episodes occur often, the growth of the char should be slower, but this has not been shown in the present study. The condition of adult char has not been shown to be affected. The reproduction of trout in the Harrsjöarna lakes has not yet been analysed. Trout populations exist and their growth is rather good, but their condition is poorer than it was in earlier years. It seems probable that all salmonid populations are affected by the poisonous effects of aluminium, but the different populations dispose the available resources very differently in acid lakes (LINDSTRÖM and ANDERSSON 1981). This was discussed more explicitly in a paper which gave a preliminary analysis of the food availability, food selection by salmonids and the secondary producion in these lakes (LINDSTRÖM *et al.* 1982). The ecology including growth of the normal char in Lakes Nedre Särnamannasjön and Stora Rösjön, and of the trout in Lakes Harrsjöarna show important differences between the lakes. Such situations may exist in other areas and thus explain the divergent results obtained from growth studies in acid lakes.

V. SUMMARY

The lakes in the Fulufjäll high mountain range are acidified, and the main management measure is liming of the lakes.

According to year-class estimates from a char population on the verge of extinction in Lake Stora Rösjön, the occurrence of poor year classes can be satisfactorily explained by an aluminium concentration above 70 µg/litre in the second half of their first year. Most of the aluminium should occur in unchelated form.

When stocking new char populations in Lake Nedre Särnamannasjön and Lake Stora Harrsjön, it was necessary to raise the pH by liming, and decrease the aluminium concentration to below 70 µg/litre in the second half of the first year of the life of the char. Establishment of the char population in Lake Stora Harrsjön was still delayed, probably due to competitive interaction by trout.

The length-weight condition of young char responded in a similar way in two of the lakes in the Fulufjäll mountain range, but the deviations which occur can probably be explained by differences between the lakes.

Fish populations in acid lakes utilize the available resources very differently.

VI. ACKNOWLEDGMENTS

Our sincere thanks are due to Ms. CATHY HILL for general comments as well as well-qualified language corrections.

VII. REFERENCES

BAKER, J. P. and C. L. SCHOFIELD. 1980. Aluminium toxicity to fish as related to acid precipitation and Adirondack surface water quality. p. 292—293. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

DICKSON, W. 1978 a. Aluminium-pH-fisk. The National Swedish Environmental Protection Board, Memorandum 1978-09-02. 40 p. (In Swedish.)

— 1978 b. Some effects of the acidification of Swedish lakes. Proc. Int. Ass. Limnol. 20: 851—856.

— 1980. Properties of acidified water. p. 75—83. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

— 1983. Liming toxicity of aluminium to fish. Vatten 39: 400—404.

ELLIOT, J. M. 1976. Body composition of brown trout (*Salmo trutta* L.) in relation to temperature and ration size. J. Anim. Ecol. 45: 273—289.

ERICHSEN JONES, J. R. 1939. The relation between the electrolytic solution pressures of the metals and their toxicity to the stickleback (*Gasterosteus aculeatus* L.) J. Exp. Biol. 16: 425—427.

— 1964. Fish and river pollution. Butterworths, London. 203 p.

LEIVESTAD, H., G. HENDREY, I. P. MUNIZ and E. SNEKVIK. 1976. Effects of acid precipitation on freshwater organisms. *In* Impact of acid precipitation on forest and freshwater ecosystem in Norway. SNSF project FR 6/76. Oslo—Ås. 87—111.

— I. P. MUNIZ and B. O. ROSSELAND. 1980. Acid stress in trout from a dilute mountain stream. p. 318—319. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

LINDSTRÖM, T. and G. ANDERSSON. 1981. Population ecology of salmonid populations on the verge of extinction in acid environments. Rep. Inst. Freshw. Res., Drottningholm 59: 81—95.

— W. DICKSON, M. HANSON and G. ANDERSSON. 1982. Low condition factor in fish of acidified lakes — an effect of insufficient food or physiological stress. Inform. Inst. Freshw. Res., Drottningholm (5). 24 p. (In Swedish with English summary.)

MUNIZ, I. P. and H. LEIVESTAD. 1980. Toxic effects of aluminium on the brown trout, *Salmo trutta* L. p. 320—321. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

NELSON, J. A. 1980. Physiological observations on developing rainbow trout, *Salmo gairdneri* (RICHARDSON) exposed to low pH and varied calcium ion concentrations. J. Fish. Biol. 20: 359—372.

NYMAN, L., J. HAMMAR and R. GYDEMO. 1981. The systematics and biology of landlocked populations of Arctic char from northern Europe. Rep. Inst. Freshw. Res., Drottningholm 59: 128—141.

PETERSON, R. H., P. G. DAYE and J. L. METCALFE. 1980. The effects of low pH on hatching of Atlantic salmon eggs. p. 328. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

RUNN, P. 1982. Effects of low pH on the development of the early stages of fish. Acta Univ. Upsaliensis 647. 85 p.

Schofield, C. L. 1977. Acid snow-melt effects on water quality and fish survival in the Adirondack mountains of New York State. Res. Rep. Project A-072-NY. 32 p.

SWEDISH STANDARD METHODS. (Svensk Standard.) 1981. SIS, Box 3295, S-103 66 Stockholm. (In Swedish.)

An Ecological Jig-Saw Puzzle:
Reconstructing Aquatic Biogeography and pH in an Acidified Region

JENS PETTER NILSSEN

Zoological Institute, University of Oslo, P. O. Box 1051, Blindern, N-0316 Oslo 3, Norway

ABSTRACT

Pelagic and littoral community structure during previous periods can be reconstructed from present acidified lakes by a thorough knowledge of the ecology of the organisms inhabiting the area to-day, field perturbation experiments and paleolimnological analyses. The results obtained can be compared with previous chemical and biological data, if available. In the present study the reconstructed chemistry and zooplankton communities for 1920—40 agreed well with samples from that period.

Plankton samples were taken from 150 lakes and paleolimnological samples from 7 lakes in a small watershed in Southern Norway which had become progressively more acidic the last 50 years. In the paleolimnological samples it became evident that the acid vulnerable *Daphnia* has disappeared the last 20—40 years, following an increase in *Bosmina*. *Daphnia* did not disappear as a consequence of increasing predation by planktivorous fish, but as a direct effect of increasing acidification. The changes in distribution of *Bosmina* morphs indicated that food availability in the surveyed lakes probably was more seasonally stable previously, because fish by selective predation stabilized the zooplankton community. Seasonal changes in phytoplankton biomass may thus be more conspicuous in acidified lakes devoid of fish than when they previously contained fish. *Daphnia* exhibited the same pattern of disappearance irrespective of the size of the lake, its catchment area or whether it was influenced by forestry, pasture or not. This suggests that the acidification process has a regional pattern probably less influenced by any major activity in the watersheds.

I. INTRODUCTION

Historical analyses of acidified lakes by using paleolimnological methods are very few, and they do not supply a definite answer as to the reasons for the alternations in the taxa. Changes in biota in acidified lakes are most often attributed directly to abiotic factors, such as pH (NILSSEN 1980 b). Acidification of freshwater involves more processes than a general lowering of pH. In many acidified lakes, biotic processes directly or indirectly induced by the altered chemical environments both seem more common, more easily detectable and more significant for the lake system: dying-out of fish and important filter-feeders (*Daphnia* spp.); increasing abundance of *Bosmina* spp., *Heterocope saliens*, *Chaoborus* spp.; changed predation pattern from fish to invertebrates; greater instability in the plankton system due to changes in species packing; increased size and thus fecundity of organisms due to lowered fish and increased invertebrate predation; decreased production-to-biomass (P/B) ratio of algae due to a greater share of slow growing Dinophyceae, probably little exploited by filter-feeders (SPRULES 1975, ROFF and KWIATOWSKI 1977, HENRIKSON and OSCARSON 1978, 1981, HENRIKSON *et al.* 1980, STENSON 1978, STENSON *et al.* 1978, ERIKSSON *et al.* 1980, NILSSEN 1980 a, NILSSON 1981, YAN *et al.* 1982, MARMOREK 1984).

Characteristic for most acidic regions is the scarcity of previous chemical and biological data. However, a combination of paleolimnology, knowledge of altered fish distribution, ecology of major invertebrate taxa, detailed field ecological studies, including enclosure experiments manipulated by artificial liming, fertilization and acidification allow for the reconstruction of aquatic biogeography, as well as pH before the major acidification period. The present study will report mainly on crustacean and diptera remains in the sediment core to reveal recent lake histories in areas thought to have undergone anthropogenic acidification. Lakes of very differing catchment area, topography and human use were chosen. The data from the core are compared with surface sediment cores where fish predation and present

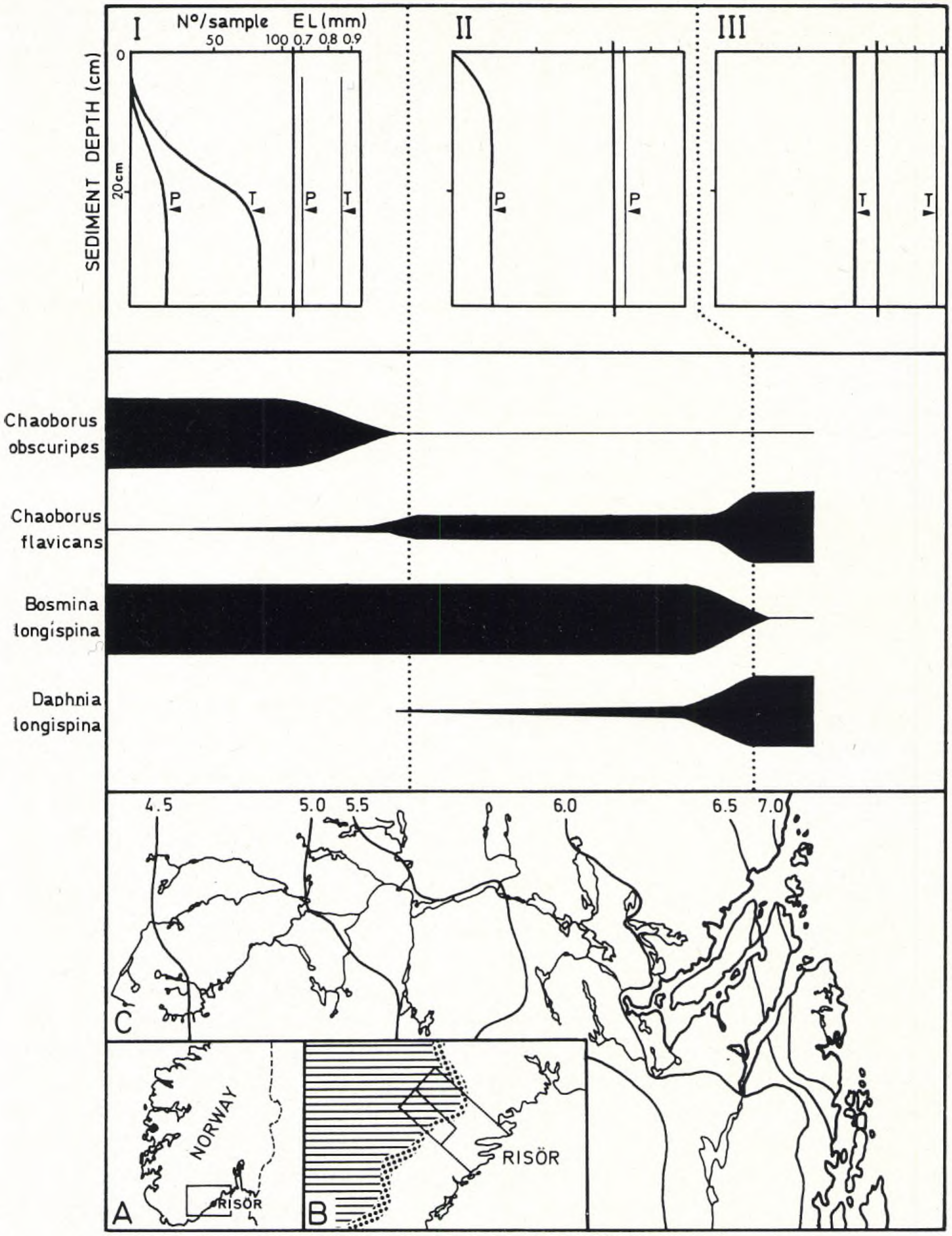


Fig. 1. Upper part: Numbers/sample of remains of *Daphnia longispina* from stratigraphic samples of lakes in the three areas I—III (semidiagrammic). EL=ephippial length. P=lakes with Eurasian perch with/without brown trout. T=lakes with only brown trout. Middle part: Regional biogeography for species treated in the text. Thickness of lines indicate abundance of species. Lower part: A=Southern Norway. B=Study area, lines indicate areas where fish has disappeared the last 30—50 years, dots where fish reproduction is no longer taking place. C=Study area in more detail. pH isoplots are based on 150 lakes at autumnal turn over.

distribution of key species in the core are known in more detail.

The methods described above will be used to reconstruct previous pH and biogeography. In the present area, earlier chemical and biological data were available to test this working strategy.

II. SITUATION

The situation of the lakes is shown in Fig. 1, an area which has undergone regional acidification the last 50 years (NILSSEN 1980 a, 1982), and receives polluted air via low pressure wind systems from West, East and Continental Europe (cf. FJERDINGSTAD and NILSSEN 1982). A decrease in pH has been recorded during the period 1965—80 in the main river draining the area (50 % of the observations decreased from above 5.9 to below 5.4 in that period), with a concurrent increase in conductivity and inorganic salts (NILSSEN 1982) Fish have disappeared from the innermost area during the last 40 years and thus changed the predation pattern from mainly vertebrate to mainly invertebrate with major changes in the pelagic zone as the ecological result.

III. MATERIAL AND METHODS

Present distribution of zooplankton taxa was assessed by using a 90 µm plankton net (32 cm opening, length 1 m) drawn twice yearly (June and September during 1974—76) from the bottom at maximum depth to the surface in 150 localities. Core samples of the Kajak type were used to reveal *Chaoborus* in localities likely to have this genus (cf. NILSSEN 1974). Stratigraphical core samples down to about 25 cm into the sediments of 7 lakes were taken from the ice during March 1981, while surface sediment samples from other lakes were available from earlier studies during the years 1977—81. Stratigraphical and surface samples were counted for remnants of *Daphnia*, *Bosmina*, *Chaoborus* and Chydoridae as described by NILSSEN (1982). Littoral sampling was done with a kick and net method and stone picking method at definite time intervals. Part of the results in Fig. 4 is also based upon other detailed studies in a restricted numbers of lakes done by the author and graduate students supervised by the author. The studies include: littoral habitat heterogeneity and life histories with and without field perturbations; life history studies of littoral species over a 4-year period; species packing and life histories in lakes dominated by fish, invertebrate predation or both; field perturbation in enclosures or involving full lakes, with and without addition of invertebrate predators; laboratory studies on cladocerans and copepods from a variety of environments related to life history strategies and relationships to the major abiotic processes associated with acidification.

IV. RESULTS AND DISCUSSION

Paleolimnological analyses

Daphnia and *Bosmina* core analyses are shown in Figs. 1—2. In the innermost lakes *Daphnia* disappears completely about 3 cm from the sediment surface, but the gradient of disappearance is very steep from 10 to 5 cm, which suggests that the process of disappearance of *Daphnia* from site 1 was a gradual process lasting for several decades (Fig. 2). Deeper than 10 cm, the number of *Daphnia* remnants does not change significantly. A sedimentation rate of ca. 2—5 mm/year is reasonable if based on studies in nearby lakes (cf. NORTON and HESS 1980). The critical years of disappearance were 20—35 years ago, which agrees with the strong increase in atmospheric pollution during 1962—65 (ODÉN 1976). In this specific field the construction of forest roads and extensive clear-cutting activities also took place in the same period. Forestry, however, cannot account for the dying-out of *Daphnia* from these lakes, since lakes outside the area of forestry show the same pattern of *Daphnia*-disappearance (NILSSEN in prep.). However, *Daphnia* started to decrease appreciably shortly after the turn of the century, suggesting that acidification of Norwegian lakes is an old process (cf. DANNEVIG 1968), probably associated with the gradual increase in fall-out over Europe following industrial build-up (cf. ODÉN 1976).

It is interesting to note that a possible increase in fish predation could not account for the disappearance of *Daphnia*, sinze the size of *Daphnia*

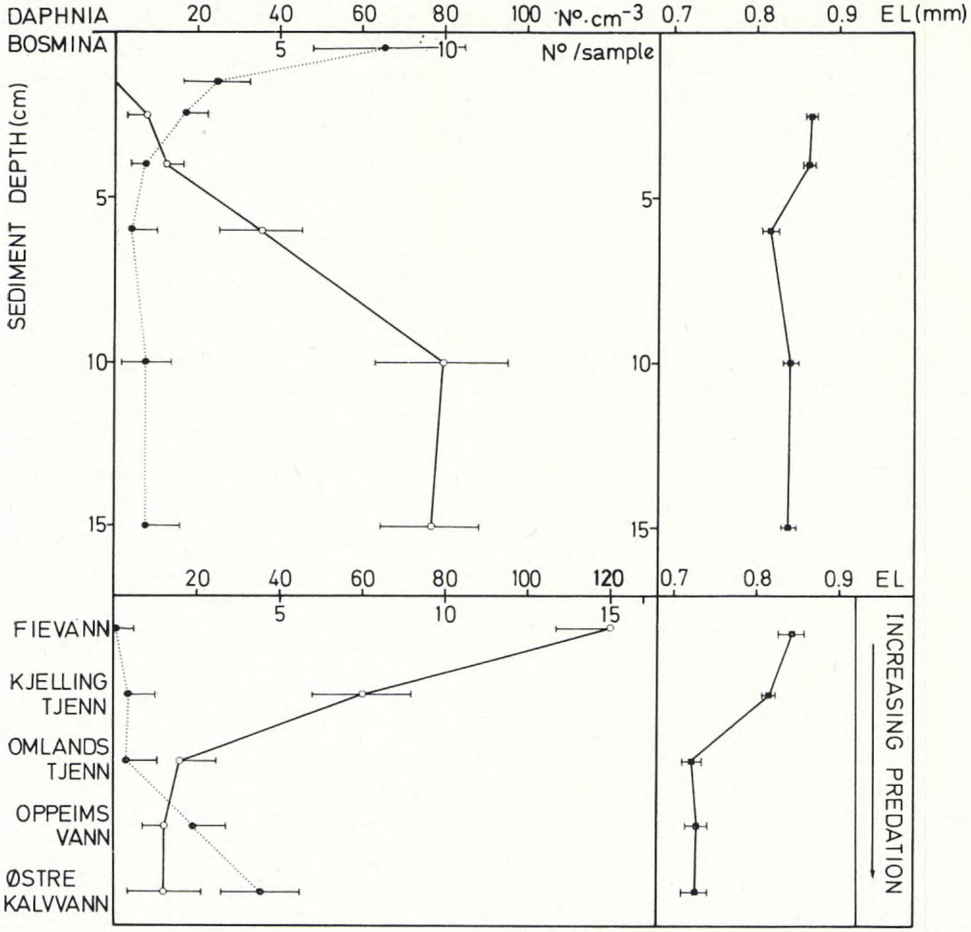


Fig. 2. Remains of *Daphnia longispina* and *Bosmina longispina* from Lake Heilandsvann (site 1) in area I (15—0 cm), Lakes Kjellingtjenn (10 cm, area I), Østre Kalvvann (10 cm, area II), Omlandstjenn (0 cm, area II), Oppheimsvann (0 cm, west Norway). Previous fish species: Heilandsvann: brown trout, now fishless; Kjellingtjenn as Heilandsvann; Østre Kalvvann: brown trout, Eurasian perch, now mainly Eurasian perch; Omlandstjenn and Oppheimsvann: *Daphnia*-consuming planktivorous brown trout. The size of remains of *Daphnia* ephippia shown to the right (EL). Mean ± S. E. shown.

increased slightly until total disappearance, which suggests that fish predation was actually decreasing (cf. NILSSEN 1978). Brown trout (*Salmo trutta*) was the only fish present in many lakes, and it disappeared about 1955—65. Simultaneously with the disappearance of *Daphnia*, *Bosmina* increased greatly in numbers (Fig. 2). It is known from present lakes that decrease of *Daphnia*, irrespective of the cause, usually results in increased numbers of *Bosmina* (NILSSEN 1978).

Lakes with relaxed fish predation had large numbers of large-sized *Daphnia* and simultaneously low numbers of *Bosmina* and vice versa (Fig. 2). The size of *Daphnia* in lakes with low abundance of brown trout near the coast was similar to site 1, while the size of *Daphnia* in lakes with heavy fish predation was smaller (Fig. 1). Therefore, *Daphnia* 50—80 years ago in site 1 experienced low predation from brown trout and it disappeared not due to fish predation, but to some factor associated with the process of acidification. If this was a direct effect of gradually lower pH or combined effects of heavy metals following acidification remains to be shown.

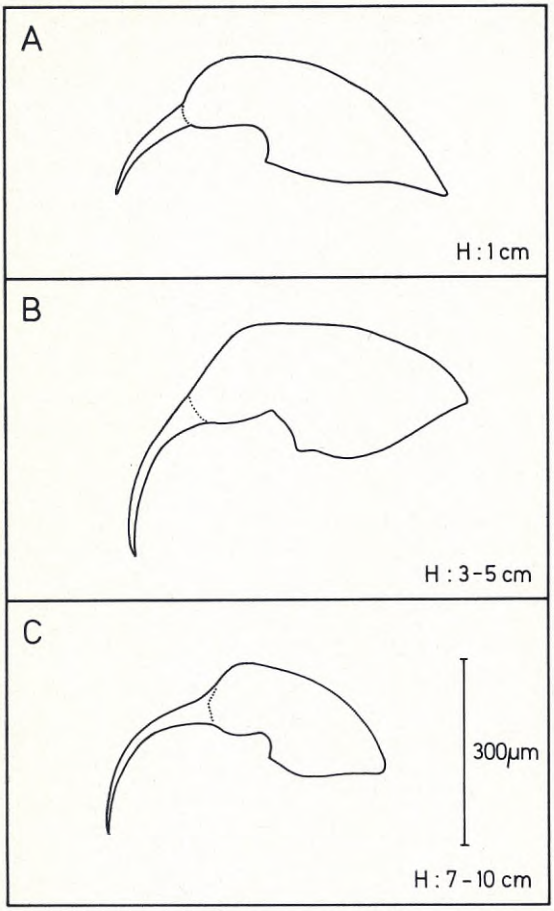


Fig. 3. Representative phenotypes of *Bosmina longispina* from Lake Heilandsvann (area I) at different depth in the core. A=morph close to *obtusirostris*. B and C=morphs close to *lacustris*.

Table 1. *Major changes in the biota of acidified lakes, including the possibility of detecting changes in the core.*

Taxa	Major change	Core detection
Fish	Selective die-out of species, starts at about pH=6.0.	No
Chaoborus flavicans	Increases strongly when fish disappear, may be excluded or suppressed by *C. obscuripes*.	Yes
C. obscuripes	Immigrate and increase strongly when fish disappear.	Yes
Heterocope saliens	Increase strongly when fish die out, competition between *C. obscuripes* and *H. saliens* not known, but probably keen.	No
Daphnia longispina	Same body-size, decreasing abundance until total disappearance.	Yes
Chydoridae	Very little known, primary suggestion: generally mean size increases due to decreased fish and increased invertebrate predation; decreasing in abundance: *Alona rectangula*, *A. costata*, increasing; *A. rustica*, *A. guttata*, more studies necessary on ecology.	Yes
Cyclopid copepods	Decreasing due to invertebrate predation (*H. saliens* and *C. obscuripes*), or factor associated directly with low pH.	No

Hydrophysiological studies indicate that the genus *Daphnia* is very sensitive directly to low pH (Potts and Fryer 1979, Nilssen et al. 1984).

Analysis of the Chydoridae from the core did not give such a clear picture as the *Daphnia-Bosmina* analysis, mainly because the ecology of Chydoridae is insufficiently known in Norway, and the interpretation lends iself too much to purely mathematical analysis (cf. Brakke 1980) and is not so much concerned with field experiments and present-day ecology of Chydoridae. Core analysis of Chydoridae did not appear as promising as did the *Daphnia-Bosmina* studies, but some general pattern could be observed. The most important were that the overall body-size of the species increased towards the sediment surface, and that some species, notably *Alona costata* and *A. rustica* increased in numbers (Table 1).

The body shape of *Bosmina* may suggest a direct relationship to predation (Kerfoot 1977, Nilssen *et al.* 1980). Representative phenotypes of *B. longispina* from a lake in site I are shown in Fig. 3. The dominant form in previous times was the *lacustris*-form of *B. longispina* which suggests that fish and invertebrate predation were high and/or food availability low (Nilssen *et al.* 1980; Nilssen in prep.). It was suggested earlier that *B. longispina* morpha *lacustris* was associated only with high fish and invertebrate predation (Nilssen *et al.* 1980), but the shape of *B. longispina* from a lake with low fish predation suggests that food availability may also play a role in the distribution of morphs, the *obtusirostris* morph usually dominating in lakes and seasons with high food availability. However, a slight change from a *lacustris* to an *obtusirostris* morph could be observed in that the length of the 2nd antennae was slightly reduced towards the sediment surface. The mean size of *B. longispina* from site 1 was larger than from sites where the species suffered heavy fish predation, supporting the hypothesis that earlier times predation patterns can also be studied by analysing sediment cores (HRBÁČEK 1969; KERFOOT 1974; NILSSEN 1978).

Breakdown of dead zooplankton is a selective process (DEEVEY 1964) and many key species in the pelagic community are not represented — species that would help greatly in the interpretation of the sediment core. Studies of the key predator *Chaoborus flavicans* indicate that this was the only chaoborid able to co-exist with fish in Norway (NILSSEN 1974). *C. flavicans* appeared to decrease greatly in anthropogenic acidic lakes when the larger, more pigmented species *C. obscuripes*, unable to co-exist with fish (STENSON 1978), was increasing greatly in numbers (see also Fig. 1 for regional biogeography of the species). It is commonly observed that the two species divide the lake between them, in that the non-migratory *C. obscuripes* is found near the shore, while the migratory *C. flavicans* occupies the deepest part of the lake (cf. STAHL 1966; HONGVE 1975).

The disappearance of fish from the present area is well known from records of the local Fishing Club (see Fig. 4). A decrease in fish abundance is usually followed by an increase in pelagic invertebrate predators, like the midges *C. flavicans*, *C. obscuripes* and the calanoid copepod *Heterocope saliens*. A concurrant decrease among the invertebrate predators, the cyclopoid copepods, is observed. This decrease was earlier suggested by NILSSEN (1980 a) as resulting from predation by *H. saliens*. One can not rule out this possibility, but the observations by EIE (1974) on the common co-occurrence of *H. saliens* and *C. scutifer* in small ponds, makes the above explanation less plausible. Moreover, an acid-vulnerable egg stage for cyclopoids in the present area has been suggested (NILSSEN in prep.).

The major changes in the biota of the pelagial in acidified lakes are summarized in Table 1 together with information that can be obtained by studying the sediment cores. The most important information is found in the *Daphnia-Bosmina-Chaoborus* complex, therefore it is very important to clarify the present ecology of these groups. Large abundance of III—IV instars of *C. obscuripes* in acidic lakes leads to very low numbers of other zooplankters. Zooplankton is only allowed to increase in numbers when the species hatches to become adults, and during its I—II instars (A. SKOV in prep.). The same key role has been suggested for *H. saliens* (BURCKHARDT 1944), but the effect of this predator is little known in the present area. It is probably not such an effective predator as *C. obscuripes* but studies in preparation indicate that it to a great extent shapes the zooplankton community in lakes also. The ecology of Chydoridae in acidic lakes is little known, but they may increase their community size, and also mean size due to increased invertebrate and decreased vertebrate predation. Habitat selection and direct relation to decreasing pH must be investigated in detail, as well as for the pelagic cyclopoids. As is evident from Table 1 it is possible to identify many changes in acidic lakes by a thorough investigation of the core, combined with ecological studies.

Reconstructing earlier biogeography and pH

The biotic structure of the pelagial of the lakes changes considerably when fish disappear (Fig. 4). More conspicuous features are the increasing biomass of these organisms which earlier were favoured prey species for fish: *H. saliens*, *C. flavicans*, *C. obscuripes*, *Glaenocorisa propinqua* and water mites (HENRIKSON and OSCARSON 1978, STENSON 1978, NILSSEN 1980 b). These important invertebrate predators may influence the ecology and seasonal abundance of other taxa and result in fewer species and lower numbers of several cladocerans, rotifers and cyclopoid copepods species, as found both in the field and in numerous enclosure experiments (NILSSEN in prep.). Some pelagic species disappear, however, as a direct effect of the acidification process, *e.g. Daphnia* spp., *Bythotrephes longimanus* and *Leptodora kindtii* (NILSSEN in prep.). Some species in the present study ex-

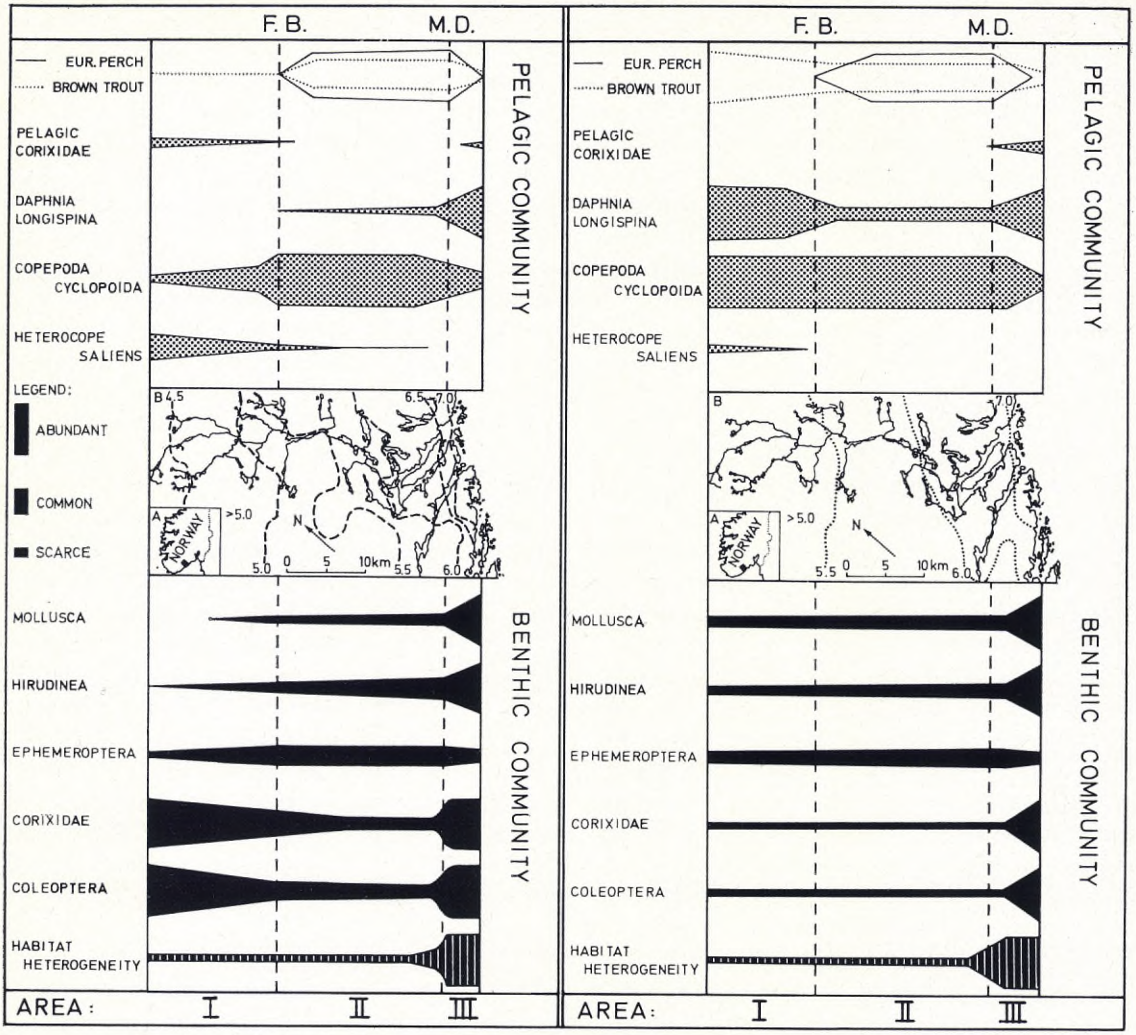


Fig. 4. Present (left) (1975) distribution of major taxa. Thickness of lines indicate abundance of taxa. pH distribution for 150 lakes at autumnal turn over. Right: reconstructed (1920—40) distribution of major taxa and pH. Legend as for 1975 samples. Areas I—III shown. Habitat heterogeneity relates to possible nos. of niches in the littoral. F. B.=friction breccia seperating two different geological region, the innermost most acidic. M.D.=lakes outside this line are situated on marine deposits and contain also large amounts of marine deposits in their catchments.

hibit a distribution suggesting a density-independent relationship to acidification: fish, gastropods and daphnids. Their physiology seem adversely affected by lake acidification (LEIVESTAD and MUNIZ 1976, POTTS and FRYER 1979).

The distribution of most organisms in the present area is probably largely determined by density-dependent factors: corixids, dytiscids, partly ephemeropterans (by fish predation); leeches (low biomass due to low prey (primarily molluscs) availability); *Daphnia* (low biomass in the mid region II due to fish predation). In the region with few available spatial refuges for prey in the littoral, all littoral taxa are probably suppressed by fish and recorded in very small numbers (Fig. 4).

Present pH and core data suggest a former distribution of pH as indicated in Fig. 4, with no changes in area III, including lakes near the coast. This agrees well with earlier measurements, since

Table 2. *Distribution of species or remnants of species from water at present and previously, surface or deep sediments for the three areas III, II and I. 100 lakes are included from the three areas for present species composition in the pelagic zone. No. of earlier samples: III (10), II (3) and I (0), sediment cores: III (4), II (4) and I (7). —: no sample 0=no specimens, x: few, xx: common, xxx: abundant, 1: no preserved, 2: C. obscuripes increases in headwater humic acidic lakes, C. flavicans decreases in lakes in the main river valley, 3: samples too small or too few to recover the species.*

Area	Environment	Period	*Daphnia longispina*	*Holopedium gibberum*	*Diaphanosoma brachyurum*	*Bosmina longispina*	Cyclopoid copepods	*Heterocope saliens*	*Chaoborus* spp. (2)	Pelagic corixids
III:	pelagic zone	1975	xxx	0	0	x	x	0	xx	x
III:	pelagic zone	1939	xxx	0	0	x	x	0	xx	0 (3)
III:	surface sediment	1970s	xxx	1	1	x	1	1	xx	3
III:	deep sediment	1920—40	xxx	1	1	x	1	1	xx	3
II:	pelagic zone	1975	x	xxx	xx	xxx	xxx	0	x	0
II:	pelagic zone	1939	x	xx	xx	xxx	xxx	0	x	0
II:	surface sediment	1970s	x	1	1	xxx	1	1	x	3
II:	deep sediment	1920—40	x	1	1	xxx	1	1	x	3
I:	pelagic zone	1975	0	xx	xx	xxx	x	xx	0	xx
I:	pelagic zone	1939	—	—	—	—	—	—	—	—
I:	surface sediment	1970s	0	1	1	xxx	1	1	0	3
I:	deep sediment	1920—40	xxx	1	1	x	1	1	xx	3

chemical data showed similar pH to present (NILSSEN 1982). Lakes in area II have experienced a lowered pH of about 0.5 pH units, while lakes in area I may have become one pH unit lower than during 1920—40. Lakes in area III have marine and lime deposits in the catchments and receive additional ions from sea aerosols. The lakes in areas I—II have poor geochemistry in their catchments and their pH have decreased considerably due to acid input and limited ability to neutralize hydronium ions.

The distribution of other organisms has been reconstructed by using data on feeding strategy of Eurasian perch and brown trout from the present area and similar environments in Scandinavia (ANDERSSON 1972, STENSON 1979, RADDUM *et al.* 1979, NILSSEN unpubl.). Important invertebrate predators in acidic lakes: *H. saliens*, *C. obscuripes* and *G. propinqua* were not recorded either from lakes containing Eurasian perch or brown trout as the fish species. The previous higher pH combined with low invertebrate and fish predation allowed both *Daphnia* spp. (corroborated from core and enclosure data) and cyclopoid copepods to be present in large numbers (indicated from present-day distribution in lakes with fish). *C. flavicans* is the only chaoborid frequently coexisting with fish in Norway (NILSSEN 1974). This species may previously have been more common in the pelagial (corroborated by core findings and present-day distribution), because the headwater lakes (area I) were previously more humic, thus providing this species with a spatial refuge in an oxygen-poor hypolimnion. Now it may be displaced, especially in shallow lakes, by its close relative *C. obscuripes*, which is commonly recorded from lakes devoid of fish. The latter species has never been recorded from the core samples, suggesting that it did not co-exist with any fish species in the present area, probably due to its conspicuous size and strong body pigmentation (STENSON 1978).

Table 2 summarizes the zooplankton and core data available from the present area, including the samples that had been stored since 1939. The observed agreements with predictions derived from Fig. 4 are striking, and thus support the validity of the present working strategy in acidification research. Moreover, littoral communities of less acidic lakes in the same area to-day strongly resemble the predicted community structure in Fig. 4.

V. ACKNOWLEDGMENTS

I thank W. KELLER, DIANA MALLEY, DAVID MARMOREK, WILLIAM NEILL, W. GARY SPRULES, NORMAN YAN, and my colleagues in the Norwegian Liming Project for informations and discussions, ROBERT A. VINTER for improving the English and INA IRENE INMAN for drawing the figures.

VI. REFERENCES

ANDERSSON, B. 1972. Food selectivity of perch (*Perca fluviatilis* L.) in acidified lakes on the west coast of Sweden. Inform. Inst. Freshw. Res., Drottningholm (17). 21 p. (In Swedish with English summary.)

BRAKKE, D. E. 1980. Atmospheric deposition in Norway during the last 300 years as recorded in SNSF lake sediment. III. Cladoceran community structure and stratigraphy. p. 272—273. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

BURCHHARDT, G. 1944. Verarmung des Planktons in kleinen Seen durch *Heterocope*. Z. Hydrol. 10: 121—124.

DANNEVIG, G. 1968. Surt vann og dødelighet på ørret. Zool. Revy. 29: 53—60. (In Norwegian.)

DEEVEY Jr., E. S. 1964. Preliminary account of fossilization in Rogers Lake. Verh. internat. Verein. Limnol. 15: 981—992.

EIE, J. A. 1974. A comparative study of the crustacean communities in forest and mountain localities in the Vassfaret area (southern Norway). Norw. J. Zool. 22: 177—205.

ERIKSSON, M. O. G., L. HENRIKSON, P. K. LARSSON, B.-I. NILSSON, H. G. NYMAN, H. G. OSCARSSON and J. A. E. STENSON. 1980. Predator-prey relations, important for the biotic changes in acidified lakes. Ambio 9: 248—249.

FJERDINGSTAD, E. and J. P. NILSSEN. 1982. Bacteriological and hydrological studies on acidic lakes in Southern Norway. Arch. Hydrobiol. Suppl. 64: 443—483.

HENRIKSON, L. and H. G. OSCARSON. 1978. Fish predation limiting the abundance and distribution of *Glaenocorisa p. propinqua* (Hemiptera). Oikos 31: 102—105.

— and H. G. OSCARSON. 1981. Corixids (Hemiptera — Heteroptera) the new top predators in acidified lakes. Verh. internat. Verein. Limnol. 21: 1616—1620

— H. G. NYMAN, H. G. OSCARSON and J. A. E. STENSON. 1980. Trophic changes, wihout changes in the external nutrient loading. Hydrobiologia 68: 257—263.

HONGVE, D. 1975. On the ecology and distribution of *Chaoborus* (Chaoboridae, Diptera) from the Upper Romerike District, south-east Norway. Norw. J. Ent. 22: 49—57.

HRBÁČEK, J. 1969. On the possibility of estimating predation pressure and nutrition level of populations of *Daphnia* (Crust., Cladoc.) from their remains in sediments. Mitt. internat. Verein. Limnol. 17: 269—274.

KERFOOT, W. C. 1974. Net accumulation rates and the history of cladoceran communities. Ecology 55: 51—61.

— 1977. Competition in cladoceran communities: the cost of evolving defence against copepod predation. Ecology 58: 303—313.

LEIVESTAD, H. and I. P. MUNIZ. 1976. Fish kill at low pH in a Norwegian river. Nature (Lond.) 259: 391—392.

MARMOREK, D. R. 1984. Changes in the temporal behaviour and size structure of plankton systems in acid lakes. p. 23—41. *In* Early biotic responses to advancing lake acidification. Ed. G. R. Hendrey. Ann Arbor Publ., Ann Arbor, USA.

NILSSEN, J. P. 1974. On the ecology and distribution of the Norwegian larvae of *Chaoborus* (Diptera, Chaoboridae). Norsk. ent. Tidsskr. 21: 37—44.

— 1978. Selective vertebrate and invertebrate predation — some paleolimnological implications. Polsk. Arch. Hydrobiol. 25: 307—320.

— 1980 a. Acidification of a small watershed in southern Norway and some characteristics of acidic aquatic environments. Int. Rev. Hydrobiol. 65: 177—207.

— 1980 b. Acidification of freshwater and limnetic organisms. Complex biotic interactions or statistics? p. 344. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås. (Abstract from poster presented at the symposium.)

— 1982. La detection de l'acidification regionale des lacs et des ses causes. Eau du Quebec 15: 335—341.

— G. HALVORSEN and J. G. MELÅEN. 1980. Seasonal divergence of *Bosmina* morphs. Int. Rev. Hydrobiol. 65: 507—516.

— T. ØSTDAHL and W. T. W. POTTS. 1984. Species replacements in acidified lakes: Physiology, predation or competition? Rep. Inst. Freshw. Res., Drottningholm 61: 148—153.

NILSSON, B. I. 1981. Susceptibility of some Odonata to fish predation. Verh. internat. Verein. Limnol. 21: 1612—1615.

NORTON, S. A. and T. HESS. 1980. Atmospheric deposition in Norway during the last 300 years as recorded in SNSF lake sediments. I. Sediment dating and chemical stratigraphy. p. 268—269. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

ODÉN, S. 1976. The acidity problem — an outline of concepts. Wat. Air, Soil Poll. 6: 137—166.

POTTS, W. T. W. and G. FRYER. 1979. The effects of pH and salt content on sodium balance in *Daphnia magna* and *Acantholeberis curvirostris* (Crustacea: Cladocera). J. Comp. Physiol. 129: 289—294.

RADDUM, G. G., J. JASTREY, B. O. ROSSELAND and I. Sevalrud. 1979. Vannteger i Sør-Norge og deres betydning som fiskeføde i vann med ulik pH. SNSF-project IR 50/79 Oslo—Ås. 41 p. (In Norwegian with English summary.)

ROFF, J. C. and R. E. KWIATKOWSKI. 1977. Zooplankton and zoobenthos communities of selected northern Ontario lakes of different acidities. Can. J. Zool. 55: 899—911.

SPRULES, W. G. 1975. Midsummer crustacean zooplankton communities in acid-stressed lakes. J. Fish. Res. Bd Can. 32: 389—395.

STAHL, J. B. 1966. Coexistence in *Chaoborus* and its ecological significance. Invest. Indiana Lakes & Streams. 7: 99—113.

STENSON, J. A. E. 1978. Differential predation by fish on two species of *Chaoborus* (Diptera, Chaoboridae). Oikos 31: 98—101.

— 1979. Predator — prey relations between fish and invertebrate prey in some forest lakes. Rep. Inst. Freshw. Res., Drottningholm 58: 166—183.

— T. BOHLIN, L. HENRIKSON, B.-I. Nilsson, H. G. NYMAN, H. G. OSCARSON and P. LARSSON. 1978. Effects of fish removal from a small lake. Verh. internat. Verein. Limnol. 20: 794—801.

YAN, N. D., C. J. LAFRANCE and G. G. HITCHIN. 1982. Planktonic fluctuations in a fertilized, acidic lake: the role of invertebrate predators. p. 137—154. *In* Acidic precipitation and fisheries impact in Northeastern North America. Eds.: T. A. Hained and R. E. Johnson. Proc. Symp. Cornell Univ. 4.

Species Replacements in Acidified Lakes: Physiology, Predation or Competition?

JENS PETTER NILSSEN,[1] TORBJØRN ØSTDAHL[1] and WILLIAM T. W. POTTS [2]

[1] Zoological Institute, University of Oslo, P. O. Box 1051, Blindern, N-0316 Oslo 3, Norway.
[2] Department of Biological Sciences, University of Lancaster, Lancaster LA1 4YQ, England.

ABSTRACT

The major processes that affect acidified lakes are changing physiological, competitive and predatory interactions. Major pelagic taxa, like fish and daphnids are physiologically strongly vulnerable to acidic environments. Copepods, water mites and *Chaoborus* showed good sodium balance in acidic waters primarily because their internal fluids are more effectively insulated from the ambient water. Life history interactions between selective predators and prey may lead to local extinction of the prey in many environments. Competitive interactions among the cladocerans are probably only partly responsible for species replacement in that group.

I. INTRODUCTION

The community structure of anthropogenic acidic lakes is different from that in other lakes in the same biogeographical region. The most striking difference is the shift in species composition. The ultimate causes of species replacements in acidic lakes are not clear, because the methods used or the details of the investigations do not usually permit precise analyses. In many earlier surveys the ambient pH was plotted on the abscissa and the numbers or biomass of all groups of plants or animals on the ordinate. This was usually interpretated as a causal relationship. Such oversimplified approaches do not give any insights into the processes associated with acidification.

The most obvious processes in acidified lakes are those with a direct physiological relationship to pH, resulting in decreased species diversity. Mollusca, Pisces and many Cladocera show such a response (Leivestad and Muniz 1976, Potts and Fryer 1979, Malley *et al.* 1982). Far more complicated than physiological events are ecological processes, such as competition and predation. As a consequence of acidification predation patterns change from mainly vertebrate (fish, salamanders) to predominantly invertebrate (Odonata, Corixidae, *Chaoborus*, *Heterocope*) predators (Eriksson *et al.* 1980, Nilssen 1980). These predation patterns can be assessed through paleolimnological evidence, or more directly through field experiments (Marmorek 1982, Yan *et al.* 1982, Nilssen *et al.*, in prep.). Evidence for competitive replacements can be found in the sediment history of the lakes (Nilssen 1984), and in the result of enclosure experiments (Wærvågen *et al.* in prep.).

The aim of this study is to report on community changes in anthropogenic acidified lakes, and to present evidence for the processes likely to cause such changes.

II. GROUPS AFFECTED BY ACIDIFICATION

The group of animals which is most likely to withstand acidification in any specific case depends on the initial composition of the fauna. Although no community interactions are identical, there is no reason to believe that acidic lakes behave differently from non-acidic ones.

The taxa most frequently affected by acidification are listed in Table 1. The groups of animals reported to be most sensitive are fishes, daphnids, molluscs, and mayflies (Almer *et al.* 1978); all these decrease rapidly in numbers or disappear in acidic lakes. On the other hand, Odonata, Corixidae, *Notonecta* and *Chaoborus* tend to increase.

Table 1. *Taxa, which vary in number during the process of acidification of lakes. Ph: physiological interaction, Pr: predatory interaction, Co: competitive interaction. Fish predation=relationship to fish predation.*

Taxa	Fish predation	Numerical change	Kind of reaction
Mollusca	Vulnerable	Disappear	Ph
Ephemeroptera	„	Many species disappear	Ph
Corixidae	„	Increase greatly	Ph
Notonecta	„	„	Ph
Odonata	„	„	Ph
Chaoboridae	„	„	Ph
Daphnia	„	Disappear	Ph
Bosmina	Partly vulnerable	Increase	Co, Pr
Diaphanosoma	„	„	Co, Pr
Holopedium	Vulnerable	„	Co, Pr
Heterocope saliens	Partly vulnerable	Increase greatly	Pr
Cyclopoid copepods		Decrease or disappear	Pr, Ph

III. PHYSIOLOGICAL INTERACTIONS

Fish and some invertebrates from strongly acidified regions are adversely affected biochemically, and under experimental conditions of low pH they show lower weight gains independent of feeding (Raddum 1979, Muniz 1981). Native brown trout (*Salmo trutta*) which were physiologically stressed showed reduced growth compared to brook trout (*Salvelinus fontinalis*) which were not being affected by the acidic water. Following transplantation the animals returned to the rate of growth appropriate to their size (Rosseland pers. comm.). Acidification alters the biochemical composition of organisms because metabolic stress results in more energy being used for maintainance.

Short-term stresses are also detectable in acidic waters, animals affected by short acid episodes were found to have temporally altered blood compositions (Leivestad and Muniz 1976).

Sodium transport (Na^{22}) is affected by the ambient concentration of H^+ (Potts and Fryer 1979, Havas and Hutchinson 1983, Havas in prep., Nilssen et al. 1983). This is evident from Fig. 1 where sodium balance for zooplankton from acidic and non-acidic sites are compared. Cladocerans are generally more adversely affected in acidic waters than are copepods (Fig. 1 A vs. 1 B). Among cladocerans, *Daphnia* spp. are more vulnerable than either *Bosmina* or *Holopedium* (Fig. 1 A). Sodium uptake was strongly depressed in roach (*Rutilus rutilus*) at low pH. Other groups investigated apparently maintained a good sodium balance (Hydracarina, *Chaoborus flavicans*, *C. obscuripes*), while other species were affected by low pH (*Daphnia pulex*, *D. magna*, *Heterocope appendiculata*) (Nilssen *et al.* 1983). Generally, sodium influx was more affected than

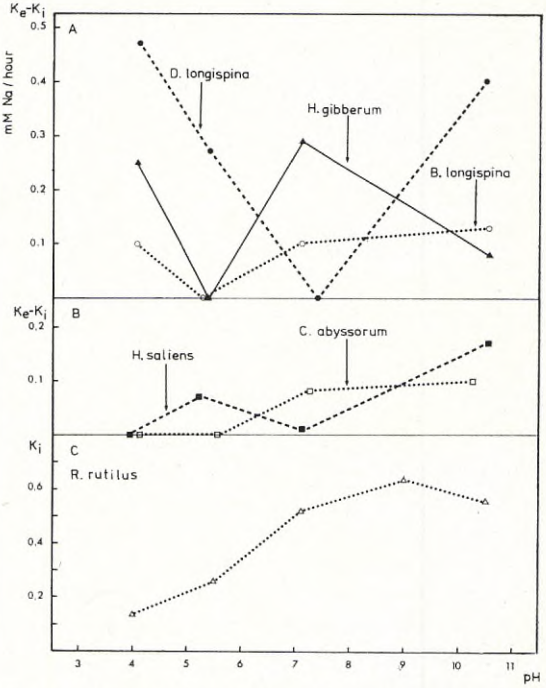


Fig. 1. A & B: Sodium balance of selected zooplankton, C: sodium uptake of roach. The method used is described in Potts and Fryer (1979) and Nilssen *et al.* (1983). Net loss: difference between efflux and influct measured by use of Na^{22}.

efflux at low pH. In an experiment with H_3THO on osmotic flux patterns we discovered the probable reason for the different reactions. Water mites and *Chaoborus* spp. were almost impermeable to water and the net osmotic flux was equivalent to 5—10 % of the total body water/*day*, in *Daphnia* it was equivalent to 20—25 % of the total body water/*hour*. Copepods were intermediate in their response and took several days to replace body water. Thus, daphnids have much more intimate contact with the ambient medium than copepods, *Chaoborus* or water mites. There was furthermore evidence, based on their Na^{22} uptake curves that *Gammarus lacustris* was much more permeable to ions than were different species of Odonata and Corixidae, while *Asellus aquaticus* occupied an intermediate position (NILSSEN *et al.* unpubl. data).

The relationship of sodium efflux to combinations of H^+, Al with and without chemical chelators was also investigated (NILSSEN *et al.* 1983), but no obvious effects were found either with *Gammarus* or *Asellus*. HAVAS (pers.comm.) reports that sodium influx was more affected than efflux with increasing concentrations of Al.

We assume that the response of animals in short-term laboratory experiments are analogous to their response in lakes undergoing acidification.

IV. COMPETITIVE INTERACTIONS

Biotic interactions during acidification are far more difficult to assess and consequently only indirect evidence is available.

In acidic lakes Dinophyceae is the predominant phytoplankton group, while Bacillariophyceae has generally a low biomass (ALMER *et al.* 1978, HENDREY *et al.* 1981). Adding lime to experimental enclosures or acidic lakes causes a shift in the algal species composition to other groups, usually those found in non-acidic lakes. Characteristically, photosynthesis seems to be partially surpressed in many acidic lakes, resulting in very low production-to-biomass ratios (KALKINGSPROSJEKTET 1983). Competitive interactions involving the phytoplankton in acidic lakes are not known.

Sediment analysis frequently shows that *Bosmina* replaces *Daphnia* as the most common Cladocera upon acidification (NILSSEN 1984). A similar relationship between species and pH was found when analysing a large number of lakes along a pH gradient (NILSSEN 1980). There is evidence that *Daphnia* is more successful than *Bosmina* under conditions of low fish predation when the pH is above 6.0, since, when introduced to enclosures they always greatly outnumbered or displaced Bosmina under these conditions. In the abscence of *Daphnia*, *Bosmina* became very numerous (WÆRVÅGEN *et al.* in prep.). The numerical relations described above was probably not due to the differential sensitivity to pH, since *Bosmina* do well over a wide pH range (Fig. 1). There is indirect evidence that other Cladocera, *e.g. Diaphanosoma* and *Holopedium*, also compete with *Daphnia*, but that *Daphnia* is more successful in conditions of low fish predation and above pH 6.0 (*e.g.* ALLAN 1973). However, an alternative hypothesis that would account for the difference of *Daphnia* at low pH would be a gradual loss in competitive ability due to metabolic stress compared with the above species. However, it was found that *Daphnia* did not decrease its reproductive ability at low pH. Those individuals which survived at lower pH levels produced broods as numerous or as frequently as unstressed daphnids, but reduced survivorship and delayed onset of reproductive maturity were observed (WALTON *et al.* 1982). This suggests that the difference of *Daphnia* is probably due to a deterioration of the environment, and not so much due to competitive interactions with the other cladoceran species.

Other taxa that are thought to interact in a competitive way in acidified lakes, are the important invertebrate predators *Chaoborus* spp. and *Heterocope saliens*. *Heterocope* develops large populations only if *Chaoborus* is not common.

V. PREDATORY INTERACTIONS

When predation patterns in acidic lakes change from a vertebrate to an invertebrate one, the impact on the total community is strongly altered, vertebrate predators consuming large and invertebrate predators small-sized prey.

Many acidic lakes are characterized by unusual groups of predators, such as pelagic corixids

(Eriksson *et al.* 1980, Henrikson and Oscarson 1981), which forms an attractive group of prey for fish in non-acidic lakes. In some Swedish lakes corixids are strong predators, shaping part of the pelagic community of lakes, and determining the abundance of many species (Henrikson and Oscarson 1981). This predatory role is played in many Norwegian lakes by the voracious species *C. obscuripes*. When this species is present in its fourth larval instar, the whole zooplankton community is greatly simplified and most prey species become rare. Zooplankton numbers increase only when this species pupates or is present in its first 1—2 developmental instars (Nilssen pers.observation). The larvae of this species consume most other zooplankton, and evidence suggests that they control zooplankton abundance (Nyberg pers. comm.). It is commonly observed that zooplankton in acidic lakes where *Chaoborus* is the major predator often exhibits unpredictable, erratic fluctuations in abundances, because when fish disappear no other predator seems to reach equilibrium with this species (Neill 1981). *Chaoborus* are also able to consume very large quantities of *Heterocope* (Nilssen *et al.* 1983). The relationship between *Chaoborus* and *Heterocope* could as well be considered a predatory one in which the role of predator and prey is determined according to the size of the developmental stages of the two species, *i.e.* small stages of *Chaoborus* are consumed by predatory stages of *Heterocope* and vice versa.

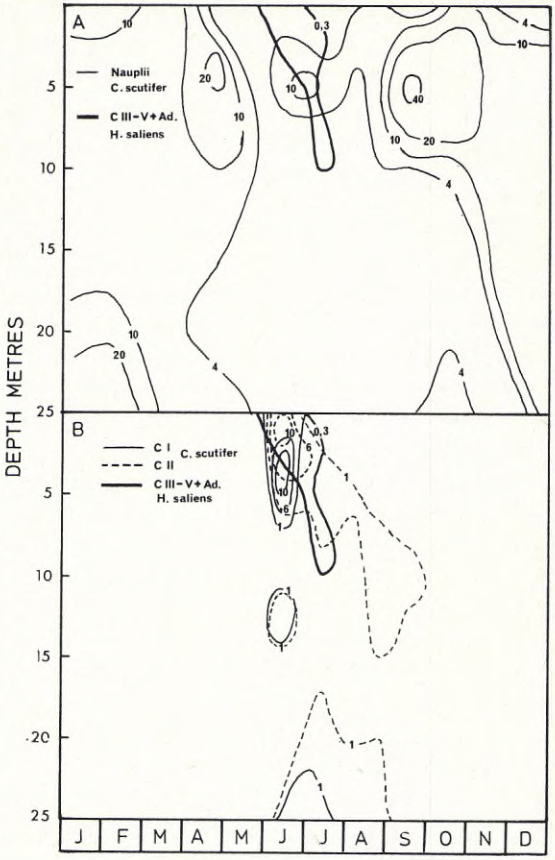


Fig. 2. Upper part: Abundance of nauplii (ind./l) of *Cyclops scutifer* during a year. Lower part: Abundance of copepodites I—II (ind./l) of *C. scutifer* from Østre Kalvvann, south Norway. In this lake planktivorous fish are still present in great abundances. On both abundances maps of *C. scutifer* is shown abundance of predatory stages (copepodites III-adults) of *Heterocope saliens* from a neighbouring lake (Heilandsvann) without fish since the 1960s.

The predatory copepod *H. saliens* which adapt to a wide range of pH (Fig. 1 B), has less influence on the zooplankton community. In ponds and in a very few lakes it may be the keystone predator (cf. Hebert and Loaring 1980), but in most lakes its major impact is probably to reduce the numbers of most prey species. It probably preys on many cyclopoid copepods (cf. Nilssen *et al.* 1983) and possibly controls the abundance of many species of this group. In acidic lakes many cyclopoid species show developmental bottle-necks, *i.e.* that the population passess through 1—2 developmental stages in a short period of time and in a restricted area of the lake. For example, we observed that *Cyclops scutifer* passed through copepod I—II in the same strata and at the same time as the predatory stages of *H. saliens* in a neighbouring lake (Fig. 2). If *H. saliens* has occurred with copepodites I—II of *C. scutifer* in the same lake, it would probably have eliminated *C. scutifer* locally. On the other hand, the nauplii of *C. scutifer* (also vulnerable to *H. saliens*' predation) was present throughout the year and at most depths (Fig. 2). Even if *H. saliens* selected this prey group, co-existence of the two species would be likely, since the prey possessed a spatial and temporal refuge from the predator. The later developmental stages of *C. scutifer* were distributed at many depths during most of the year, thus permitting co-existence of predator and prey. The simultaneous presence of *H. saliens* and *C. scutifer* is very rare in acidic lakes, indicating that *H. saliens* may usually eliminate *C. scutifer* by consuming its »bottle-neck stages» according to the mechanism described above. Such special copepod interactions are common in nature (cf. NILSSEN 1978).

To summarize, two constrasting invertebrate groups take over the predatory role of fish in acidic lakes. The first group include non-selective keystone predators, (*e.g. Chaoborus* spp.), where prey abundance is restricted to periods when the predators do not possess predatory stages or some developmental stages temporarily leave the habitat. With such predators, large erratic fluctuations in prey abundances will occur. In the presence of stabilizing fish predators, these invertebrates are usually predator controlled, and prey numbers are more stable. The second group include more selective predators (*e.g. Heterocope*) which exert a smaller influence on the community, and the co-existence of predator and prey is common.

VI. CONCLUSION

The relationship of different taxa to acidification is summarized in Table 1. Some taxa show a complicated pattern in that a combination of processes is found. Both daphnids and molluscs are attractive prey to fish, and their abundances are therefore suppressed when fish are present. Simultaneously, however, they are physiologically strongly vulnerable to acidic environments. More work is needed, especially on competitive and predatory interactions, to assess the major processes during acidification.

VII. ACKNOWLEDGMENTS

We are grateful for discussions with MAGDA HAVAS, Toronto, Canada, LENNART HENRIKSON and HANS G. OSCARSON, Gothenburg and PER NYBERG, Stockholm, Sweden. MEL WHITESIDE, University of Tennessee, USA gave important comments that improved the paper, and WENCHE RYDNINGEN kindly typed the manus. This paper was partly supported by the Norwegian Liming Project 1980—1984.

VIII. REFERENCES

ALLAN, J. D., 1973. Competition and the relative abundance of two cladocerans. Ecology 54: 484—498.

ALMER, B., W. DICKSON, C. EKSTRÖM and E. HÖRNSTRÖM. 1978. Sulfur pollution and the aquatic ecosystem. p. 271—311. *In* Sulfur in the environment: Part II. Ecological impacts. Ed.: J. O. Nriagu. J. Wiley & Sons, New York.

ERIKSSON, M. O. G., L. HENRIKSON, P. K. LARSSON, B.-I. NILSSON, H. G. NYMAN, H. G. OSCARSON and J. A. E. STENSON. 1980. Predatory-prey relations, important for the biotic changes in acidified lakes. Ambio 9: 248—249.

HAVAS, M. and T. C. Hutchinson. 1983. Effect of low pH on the chemical composition of aquatic invertebrates from tundra ponds at the Smoking Hills, N.W.T., Canada. Can. J. Zool. 61: 241—249.

HEBERT, P. D. N. and J. M. Loaring. 1980. Selective predation and the species composition of arctic ponds. Can. J. Zool. 58: 422—427.

HENDREY, G. R., N. D. YAN and K. J. BAUMGARTNER. 1981. Responses of freshwater plants and invertebrates to acidification. p. 457—466. *In* EPA/OBCD International Symposium for Inland Waters and Lake Restoration, Portland, Maine, USA.

HENRIKSON, L. and H. G. Oscarson. 1981. Corixids (Hemiptera-Heteroptera), the new top predators in acidified lakes. Verh. internat. Verein. Limnol. 21: 1616—1620.

KALKINGSPROJEKTET. 1983. Årsrapport 1980/81. Kalkingsprosjektet i Gjerstad, Aust-Agder. Kalkingsprosjektet, Miljøverndept. Norge Rapp. 6/83: 1—101. (In Norwegian.)

LEIVESTAD, H. and I. P. MUNIZ. 1976. Fish kill at low pH in a Norwegian river. Nature, Lond. 259: 391—392.

MALLEY, D. F., D. L. FINDLAY and P. S. S. CHANG. 1982. Ecological effects of acid precipitation on zooplankton. p. 297—327. *In* Acid precipitation — effects on ecological systems. Ed.: F. M. D'Itri. Ann Arbor Publ., Ann Arbor, USA.

MARMOREK, D. R. 1982. Changes in the temporal behaviour and size structure of plankton systems in acid lakes. p. 23—41. *In* Early biotic responces to advancing lake acidification. Ed.: G. R. Hendrey. Ann Arbor Publ., Ann Arbor, USA.

MUNIZ, I. P. 1981. Acidification — effects on aquatic organisms. p. A101—A123. *In* Beyond the energy crisis — opportunity and challenge. Eds.: R. A. Fazzolare and C. B. Smith. Pergamon Press, Oxford & New York.

NEILL, W. E. 1981. Impact of *Chaoborus* predation upon the structure and dynamics of a crustacean zooplankton community. Oecologia (Berl.) 48: 164—177.

NILSSEN, J. P. 1978. On the evolution and ecology of life histories of limnetic cyclopoid copepods. Mem. Ist. Ital. Idrobiol. 36: 193—214.

— 1980. Acidification of small watershed in southern Norway and some characteristics of acidic aquatic environments. Int. Rev. Hydrobiol. 65: 177—207.

— 1984. An ecological jig-saw puzzle: reconstructing aquatic biogeography and pH in an acidified region. Rep. Inst. Freshw. Res., Drottningholm 61: 138—147.

— T. ØSTERDAHL and W. T. W. POTTS. 1983. Physiologi til zooplankton under forsurning og kalking: et pilotstudium med radioisotoper. Kalkingsprosjektet, Miljøvernsdept. Norge Rapp. 7/83: 1—31. (Mimeographed in Norwegian.)

POTTS, W. T. W. and G. FRYER. 1979. The effects of pH and salt content on sodium balance in *Daphnia magna* and *Acantholeberis curvirostris*. J. comp. Physiol. 129: 289—294.

RADDUM, G. G. 1979. Effects on low pH on insect larvae. SNSF-project IR 45/79. Oslo—Ås. 58 p. (In Norwegian with English summary.)

WALTON, W. E., S. M. COMPTON, J. D. ALLAN and R. G. DANIELS. 1982. The effect of acid stress on survivorship and reproduction of *Daphnia pulex* (Crustacea: Cladocera). Can. J. Zool. 60: 573—579.

YAN, N. D., C. J. LAFRANCE and G. G. HITCHIN. 1982. Planktonic fluctuations in a fertilized, acidic lake: the role of invertebrate predators. p. 137—154. *In* Acidic precipitation and fisheries impact in Northeastern North America. Eds.: T. A. Hained and R. E. Johnson. Proc. Symp. Cornell Univ. 4.

Impact of Chaoborus Predation on Planktonic Crustacean Communities in Some Acidified and Limed Forest Lakes in Sweden

PER NYBERG

Institute of Freshwater Research, S-170 11 Drottningholm, Sweden

ABSTRACT

Crustacean planktonic communities have been studied in five acidified forest lakes. Lakes Blanksjön, Iglafallssjön and Vibollsjön were devoid of fish at the start of the study in 1977. Lakes Blanksjön and Iglafallssjön were limed in 1978 and were stocked with brown trout (*Salmo trutta*) in 1979. Lakes Svartsjön and Långsjön were inhabited by sparse and nonreproducing populations of perch *(Perca fluviatilis)* in 1976 and were practically devoid of fish in 1980. In the lakes barren of fish, copepods, although in low numbers, dominated the crustacean community, while cladocerans also occurred in the lakes with populations of perch.

No qualitative changes were noted in the crustacean communities of Lakes Blanksjön and Iglafallssjön during the first years after liming. After their introduction, trout almost eliminated *Chaoborus*. Consequently, *Bosmina* sp., *Diaphanosoma brachyurum, Ceriodaphnia quadrangula, Holopedium gibberum* and *Daphnia longispina* started to appear. A few years later cladocerans dominated the crustacean community. In Lake Vibollsjön, only single cladocerans were observed during the period 1977—83 and the population of *Chaoborus* was very dense. In L. Svartsjön only a few copepods were found in 1980 together with numerous chaoborids. In the same year L. Långsjön was practically devoid of crustaceans except for a brief expansion of *Ceriodaphnia quadrangula* and *Chaoborus* was abundant. It seems that *Chaoborus* is able to eradicate herbivorous cladocerans in lakes devoid of other invertebrate predators and fish.

I. INTRODUCTION

Planktonic crustacean communities are exposed to strong predation by most fish populations. This results in altered size distribution within zooplankton populations and vulnerable species are often eradicated from the community (BROOKS and DODSON 1965, GALBRAITH 1967, NILSSON and PEJLER 1973). Besides fish, pelagic invertebrates, *e.g. Mysis relicta* (LASENBY and FÜRST 1981) and *Chaoborus*, may exert pronounced predation pressure on planktonic crustaceans. Predation by species of the genus *Chaoborus* has been shown to be responsible for a considerable part of the mortality in zooplankton populations (KAJAK and RANKE-RYBICKA 1970, FEDORENKO 1975, GLIWICZ *et al.* 1978, KAJAK and RYBAK 1979, LYNCH 1979, NEILL 1981).

Species of *Chaoborus* with a pronounced pelagic lifestyle, *e.g. Chaoborus americanus, C. trivittatus* and *C. obscuripes* are extremely susceptible to predation by fish (NORTHCOTE *et al.* 1978, STENSON 1978), and are never found in lakes with dense fish populations. Even *C. flavicans*, which displays migratory behaviour and spends the daylight hours in the sediment (GOLDSPINK and SCOTT 1971), is highly susceptible (STENSON 1978). This implies that the abundance of chaoborids and hence the significance of the predation on zooplankton by chaoborids is most pronounced in lakes which have sparse populations of fish or are devoid of fish.

The recent acidification of inland waters in Scandinavia and North America has led to the decline or eradication of many fish populations (SCHOFIELD 1976, HARVEY 1980, SEVALRUD *et al.* 1980, JOHANSSON and NYBERG 1981). Another feature of acidification is a decline in the number of species of planktonic crustaceans (ALMER *et al.* 1978, HOBAEK and RADDUM 1980). This effect may be attributed to the toxic effect of the water on certain species and to altered predator-prey interactions after the decline or eradication of fish (ERIKSSON *et al.* 1980). A few species, *e.g.* species of *Daphnia*, are sensitive to low pH-values, but many other species of Cladocera are frequently found in acid lakes which still contain fish (JOHANSSON and NYBERG 1981). Moreover, changes similar to those in acidified lakes have been observed after an experimental eradication of fish in a non-acid lake (STENSON *et al.* 1978).

Intensive predation by new top predators such as chaoborids in lakes devoid of fish may thus be responsible for many of the effects observed in zooplankton communities in acidified lakes.

The liming of acidified lakes has been shown to result in an increase in the number of species and the biomass of phytoplankton (ERIKSSON *et al.* 1983), as well as an increase of chlorophyll a (HENRIKSON *et al.* 1984) and primary production (LARSSON unpubl.). In lakes devoid of fish, these improved food conditions for planktonic herbivores have not been followed by a corresponding expansion of the zooplankton community (ERIKSSON *et al.* 1983, HENRIKSON *et al.* 1984).

This study was performed as a part of a project dealing with the ecological effects of the lime treatment of acidified lakes and rivers in Sweden. Due to the minor effects observed in the zooplankton communities after the liming of some lakes, special attention was paid to the possible significance of predation by *Chaoborus flavicans* in particular, as this species was found to increase after lime treatment. Similar patterns, *i.e.* increased abundances of invertebrate predators and small changes in the zooplankton community after liming, were also noted by HENRIKSON and OSCARSON (1984) and HENRIKSON *et al.* (1984).

II. MATERIAL AND METHODS

Water samples for chemical analyses were taken at three depths in each lake and analysed according to the standards of the NATIONAL SWEDISH ENVIRONMENT PROTECTION BOARD (1981).

Four zooplankton samples, each with a volume of 1.6 l, were taken with a Ruttner sampler at every second metre down to a depth of 6 m and filtered through a 150 μm mesh. The four samples from each depth were pooled and preserved in Lugol's solution, and all individuals were counted under a stereo microscope. Monthly samples were taken from May—October and during most years additional sampling occurred 2—3 times from November—April.

As only a few length measurements were made for the different species, the crustacean planktonic biomass has been calculated according to the individual fresh weights given in Table 1 (GRÖNBERG 1973, JOHANSSON pers. comm.).

Table 1. *Individual fresh weights used in calculations of crustacean biomass.*

Species	Individual weight (μg)
Diaphanosoma brachyurum	30
Holopedium gibberum	50
Daphnia longispina	50
Ceriodaphnia quadrangula	15
Bosmina sp.	15
Chydorus sphaericus	10
Polyphemus pediculus	100
Cyclops spp. (cop., ad.)	20
Eudiaptomus gracilis (cop., ad.)	20
nauplii	1

The chaoborid populations were sampled regularly at the same intervals as the crustaceans, by one single net-haul from the bottom to the surface in the deepest part of each lake. The area of the net was 830 cm^2 and the mesh size was 150 μm. This sample may be regarded as providing a rough estimate of the abundance of pelagic chaoborids in the deepest part of the lakes. Moreover, no distinction has been made between *Chaoborus flavicans* and *C. obscuripes* in these samples. The abundance of chaoborids has been expressed either as ind/m^3 on each sampling occasion or as the average value for 5—6 monthly samples during May—October.

Benthic samples were taken in December 1977, and from 1978—80 parallel benthic and pelagic samples were taken in Lakes Blanksjön and Iglafallssjön in April and December. Two benthic samples were taken with an Ekman grab at a depth of 1 and 2 metres respectively, and thereafter at every second metre down to the maximum depth in each lake. The material was sieved through a 0.6 mm mesh. Pelagic samples were taken at the same time by vertical net-hauls from the bottom to the surface at depths of 4, 8 and 14 m. The area of the net was 0.26 m^2 and the mesh size was 0.5 mm. At these three depths 5, 3, 2 and 5, 3, 3 hauls were taken at randomly chosen places in Lakes Blanksjön and Iglafallssjön respectively. The filtered volume has been calculated to be 19.4 and 20.4 m^3 respectively on each sampling occasion. These samples may be regarded as more quantitative and the abundance has been expressed as ind/m^2 for the entire lake.

When calculating the biomass of *Chaoborus* *flavicans*, the individual fresh weight was measured monthly during July—September 1977 and June—September 1978—80. The monthly individual weight was not found to differ between the different years, so the same monthly value has been used for the entire period. The value for the number of individuals in June of these years was based on the abundance in April and the value for July—September was based on the abundance in December, with the following exception. The value for the number of *Chaoborus* in July—September 1977 was based on the abundance in April 1978, (as no pelagic samples were taken in December 1977).

The calculation of food consumption of instars 3 and 4 of *Chaoborus flavicans* has been performed according to KAJAK and RYBAK (1979), and the number of feeding hours per day has been assumed to be 5.5, 8.0, 14.0 and 12.5 in June, July, August and September respectively (*op. cit.*). Because of the small amount of available prey in the lakes studied, the hourly ration of *Chaoborus flavicans* while feeding has been assumed to be 0.5 % of the animals' fresh weight (*op. cit.*).

Test fishing was carried out monthly during the ice-free season from August 1979 and October 1979 onwards in Lakes Blanksjön and Iglafallssjön respectively. On each occasion 4—17 brown trout (*Salmo trutta*) were caught and the amount of *Chaoborus* spp. in the stomach contents was expressed as mean volumetrical percentage.

III. LAKE CHARACTERISTICS

The lakes, which are all situated in forested areas (Fig. 1), are fairly small (1.6—11.5 ha) and the maximum depth varies between 7 and 15 metres (Table 2). The lakes were all seriously affected by acid deposition and the pH-values were within the range of 4.7—5.2 in Lakes Blanksjön, Iglafallssjön and Vibollsjön in October 1977 and March 1978 (Table 3). Lakes Blanksjön and Iglafallssjön were treated with lime ($CaCO_3$) in May 1978 and the treatment was repeated in May 1981 and 1983. Lake Vibollsjön has not been limed but it was affected by runoff from limed Lake Iglafallssjön. After the liming treatment the pH increased and it has since then been 5.5—7.1 in Lakes Blanksjön and Iglafallssjön, while the pH-increase was much smaller in Lake Vibollsjön (Table 3). The lakes had low concentrations of plant nutrients during the course of the investigation and no remarkable changes in the concentration of Al have been noted (Table 3).

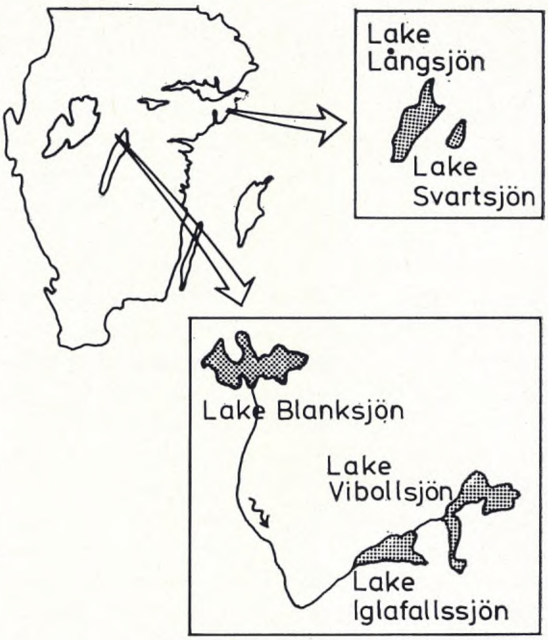


Fig. 1. Location of the lakes studied.

Table 2. *Some morphometric characteristics of the lakes studied.*

	Surface area (ha)	Volume ($m^3 \cdot 10^6$)	Maximum depth (m)	Mean depth (m)
L. Blanksjön	11.5	0.51	15.0	4.4
L. Iglafallssjön	6.1	0.34	14.0	5.6
L. Vibollsjön	10.0	0.25	7.0	2.5
L. Svartsjön	1.6	0.06	7.0	3.6
L. Långsjön	8.0	0.39	11.0	5.0

Table 3. *Some physical-chemical parameters in the lakes studied. Volume-weighted averages for the entire lake.*

Date/Parameter	14.10–77	22.3–78	8.10–78	4.5–79	23.10–79	26.4–80	24.10–80	11.5–81	19.10–81	11.5–82	15.10–82	12.5–83
L. Blanksjön												
pH	5.2	4.8	6.3	6.2	6.4	5.8	6.0	6.4	6.9	6.6	7.1	6.3
Alkalinity (meq l^{-1})	0	0	0.04	0.05	0.05	0.04	0.03	0.06	0.14	0.10	0.13	0.07
Colour (mg Pt l^{-1})	19	33	39	45	38	41	40	40	40	43	17	34
Conductivity (mSm^{-1})	3.9	4.3	4.2	4.3	4.2	4.2	4.1	4.2	5.2	5.1	5.5	4.7
Total N (mg l^{-1})	0.49	0.70	—	0.41	0.51	0.51	0.69	0.42	0.35	0.49	0.50	—
Total P (µg l^{-1})	4	5	—	13	6	6	10	5	4	5	9	—
Al (µg l^{-1})	189	274	165	172	—	—	159	179	106	229	—	159
L. Iglafallssjön												
pH	4.9	4.7	6.1	5.9	6.3	5.5	5.6	6.2	6.1	6.0	6.2	5.6
Alkalinity	0	0	0.03	0.02	0.03	0.01	0.02	0.04	0.05	0.03	0.04	0.01
Colour	55	65	63	74	67	70	87	70	68	61	35	35
Conductivity	3.8	4.2	4.0	4.0	3.8	3.8	3.7	3.9	3.6	4.0	3.9	4.2
Total N	0.37	0.57	—	0.28	0.42	0.39	0.49	0.34	0.36	0.41	0.48	—
Total P	5	6	—	10	11	7	8	6	5	5	10	—
Al	345	398	307	240	—	—	299	254	201	268	—	280
L. Vibollsjön												
pH	4.9	4.8	5.0	5.1	5.2	5.1	5.0	5.3	5.4	5.2	5.5	5.1
Alkalinity	0	0	0	0	0	0	0	< 0.01	< 0.01	< 0.01	< 0.01	< 0.01
Colour	50	150	80	90	118	91	118	75	170	93	124	45
Conductivity	3.7	4.4	3.7	3.5	3.5	5.3	3.6	3.3	3.3	3.5	3.7	4.0
Total N	0.39	0.69	—	0.40	0.43	0.43	0.59	0.34	0.56	0.34	0.47	—
Total P	7	7	—	15	11	10	12	6	15	6	15	—
Al	236	471	385	340	—	—	349	229	274	254	—	275

Date/Parameter	29.6–76	23.8–76	21.10–76	3.5–77	5.6–80	19.8–80	5.11–80
L. Svartsjön							
pH	4.8	4.7	4.7	4.6	4.7	4.9	4.6
Colour	30	16	5	75	72	59	84
Conductivity	3.1	3.4	3.6	4.1	—	—	3.8
Total N	—	—	—	0.33	—	—	0.73
Total P	—	—	8	3	—	—	4
Al	—	—	146	253	—	—	240
L. Långsjön							
pH	4.8	4.7	4.8	4.4	4.6	4.9	4.5
Colour	80	62	60	100	155	122	143
Conductivity	3.6	4.0	4.3	4.5	4.5	—	3.4
Total N	—	—	—	0.38	—	—	0.71
Total P	—	—	15	3	—	—	13
Al	—	—	320	352	—	—	371

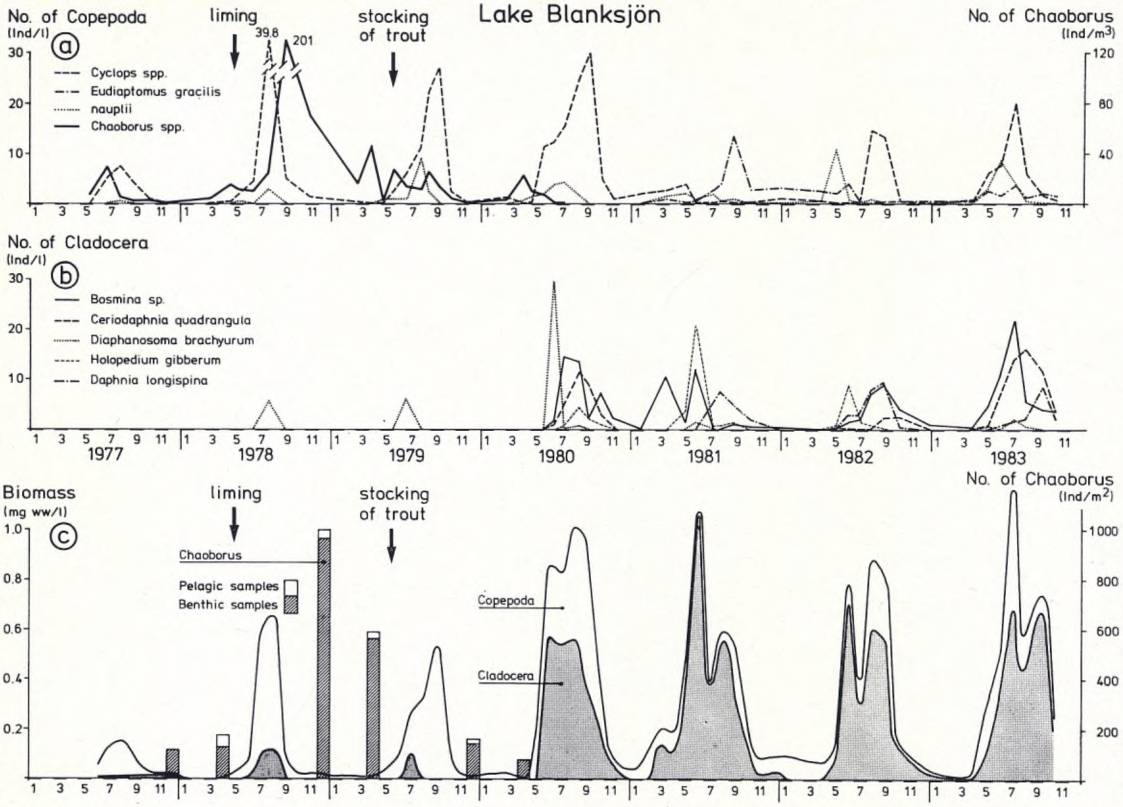


Fig. 2. (a) Abundance of Copepoda and *Chaoborus,* (b) abundance of Cladocera, and (c) crustacean biomass and *Chaoborus* density in benthic and pelagic samples in Lake Blanksjön 1977—83. (Volume-weighted averages for the water layer 0—7 m.)

The pH-values in Lakes Svartsjön and Långsjön were very low (4.4—4.9) during the entire period.

As mentioned earlier, Lakes Blanksjön, Iglafallssjön and Vibollsjön were all devoid of fish in 1977. In May 1979 3,000 yearlings of brown trout (*Salmo trutta*) were stocked in Lake Blanksjön and 5,000 fry with absorbed yolk-sacs of the same species were stocked in Lake Iglafallssjön. Lake Vibollsjön has, however, been free of fish during the entire period. Lakes Svartsjön and Långsjön were inhabited by very sparse and non-reproducing populations of perch (*Perca fluviatilis*) in 1976 and may be regarded as being almost devoid of fish during 1980.

IV. RESULTS

During 1977, before liming, pelagic crustacean abundance was very low in Lake Blanksjön. The maximum value observed was 7.5 *Cyclops* spp./l in August and very few nauplii were found (Fig. 2 a). The number of nauplii has, however, been underestimated in all the lakes because of the coarse mesh size (150 µm) of the nets used. The very small biomass (< 0.15 mg ww/l) was dominated by copepods and the extremely small biomass of cladocerans was made up of a few *Chydorus sphaericus* (Fig. 2 c). In July—August of the summer after lime treatment, the number of copepods and nauplii suddenly increased several-fold compared to the previous year, but cladocerans were still missing, except for a few *Diaphanosoma brachyurum* (Figs. 2 a, 2 b). At the same time the new generations of *Chaoborus flavicans* and *C. obscuripes* became extremely abundant in the net-hauls and the peak value was 201 ind/m³ (Fig. 2 a). Of the two species, *C. flavicans* was the more dominant and *C. obscuripes* never exceeded 10 per cent of the abundance of *C. flavi-*

cans. As a consequence of the increased density of *Cyclops* spp., crustacean biomass increased, but only for a very short period (Fig. 2 c). As can be seen in the same figure, *Chaoborus* was very numerous in December the same year (1000 ind/m²) and in April 1979 chaoborids also occurred in high numbers (590 ind/m²) (Fig. 2 c). The composition of the crustacean community was about the same during 1979 and the biomass was of the same order as the previous year (Fig. 2 c).

The stocking of brown trout in May 1979 resulted in intensive predation on *Chaoborus* spp. In Lake Blanksjön trout fed on chaoborids to a very high degree during late summer 1979 and in the following spring, just before and during the emergence of *Chaoborus* spp. (Fig. 3). The brown trout had actually already eliminated most of the populations of *Chaoborus flavicans* and *C. obscuripes* in 1979 as can be seen from both the pelagic samples (Fig. 2 a) and the quantitative samples in December 1979 and April 1980 (Fig. 2 c). Since June 1980 only single chaoborids have been found in the stomachs of trout. Planktonic crustaceans were generally never found in the stomachs of the trout. In July 1981, however, 12 per cent of the diet was composed of *Daphnia longispina*.

During the summer of 1980, the composition of the zooplankton in Lake Blanksjön changed drastically. *Bosmina* sp., *Ceriodaphnia quadrangula* and *Holopedium gibberum* started to appear and *Diaphanosoma brachyurum* occurred in high numbers in June. During 1981 and 1982 the cladoceran community was dominated by *Bosmina* sp., *Holopedium gibberum* and *Daphnia longispina*, and in 1983 *Ceriodaphnia quadrangula* and *Bosmina* sp. were the most abundant species (Fig. 2 b). The earlier dominance of *Cyclops* spp. weakened in 1981 and instead *Eudiaptomus gracilis* spp. was the most numerous copepod (Fig. 2 a). *Chaoborus* has not been found in any net-hauls since July 1980 (Fig. 2 a) but it does occur in low numbers in the deeper parts of the lake (NYBERG unpubl.).

As a result of the increased number of cladocerans, the crustacean biomass nearly doubled during 1980 compared to previous years and reached a peak value of about 1.0 mg/l in August. Apart from some variation between different years, the crustacean biomass has been fairly constant during 1980—83 and has been dominated by cladocerans (Fig. 2 c).

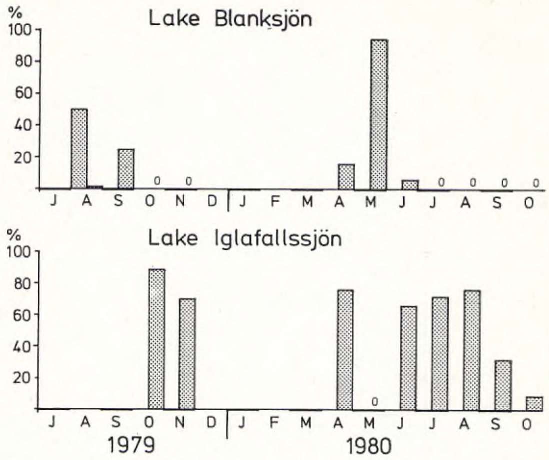


Fig. 3. Predation on *Chaoborus* by brown trout in Lakes Blanksjön and Iglafallssjön 1979—80. (Mean volumetrical percentage.)

The structure of the crustacean community in Lake Iglafallssjön before liming (1977) was about the same as in Lake Blanksjön, with the sole exception that *Eudiaptomus gracilis* was the dominant species. Chaoborids were, however, more numerous in pelagic samples (Fig. 4 a) and consisted mostly of *Chaoborus flavicans*, even in Lake Iglafallssjön.

After liming, the existing copepods *i.e. E. gracilis*, *Cyclops* spp. and nauplii increased compared to the previous year, but cladocerans were represented only by *Diaphanosoma brachyurum* during the years 1978—80 (Figs. 4 a, 4 b). During the same period the biomass increased from ca 0.14 mg/l in 1977 to 0.28—0.57 mg/l, and was dominated by copepods, especially during 1978 and 1979 (Fig. 4 c). As can be seen, chaoborids increased in the pelagic samples after liming and were numerous during all seasons of the year (Fig. 4 a). In the quantitative samples, *Chaoborus* reached a maximum density (ca 670 ind/m²) in December 1978.

In this lake, stocked brown trout fed even more intensively on chaoborids, and on most sampling occasions their diet consisted to 60—80 per cent of chaoborids until September 1980 (Fig. 3). No

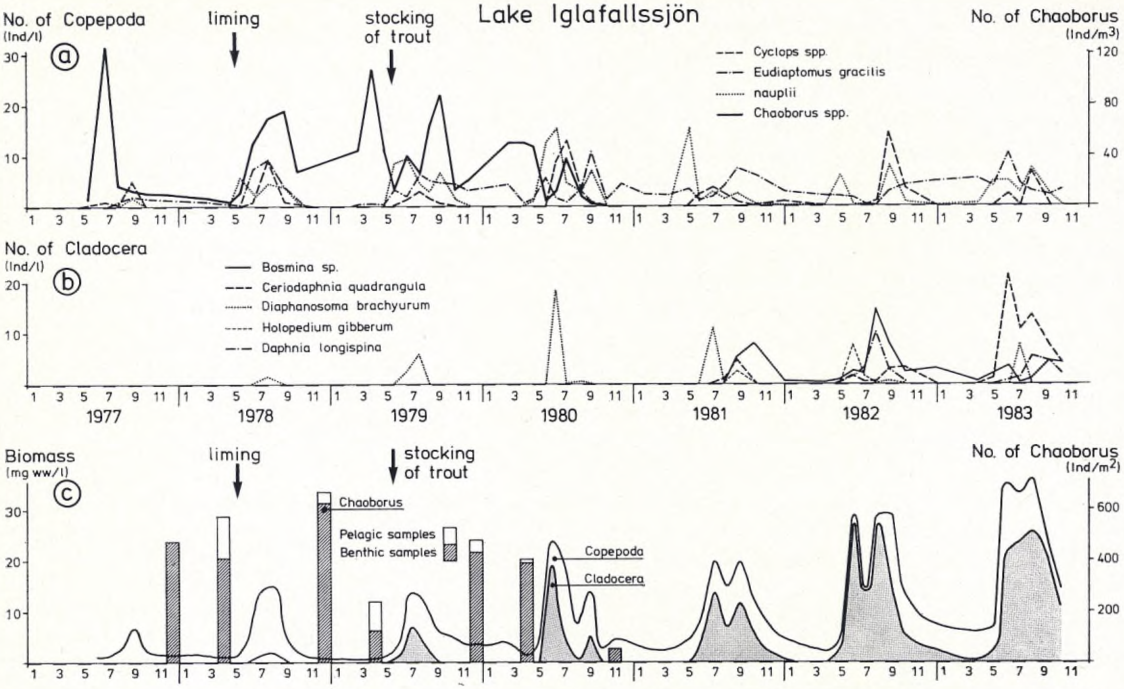


Fig. 4. (a) Abundance of Copepoda and *Chaoborus*, (b) abundance of Cladocera and (c) crustacean biomass and *Chaoborus* density in benthic and pelagic samples in Lake Iglafallssjön 1977—83. (Volume-weighted averages for the water layer 0—7 m.

predation on planktonic crustaceans by the trout was found in 1979—80. Chaoborids occurred in samples during the whole summer of 1980. Although it has not been found in pelagic samples since the end of the summer of 1980 (Fig. 4 a), *Chaoborus flavicans* was still found in benthic samples in December the same year (Fig. 4 c).

After the extreme reduction in numbers of *Chaoborus*, the crustacean community developed in a manner similar to that in Lake Blanksjön. In 1980 *Diaphanosoma brachyurum, Ceriodaphnia quadrangula, Bosmina* sp. and *Holopedium gibberum* were found in the samples, and during 1982 and 1983 *Daphnia longispina* was also common (Fig. 4 b). Copepods were, however, equally abundant during the whole period (Fig. 4 a). Due to the increase in cladocerans, the biomass of crustaceans increased slowly and reached maximum values (0.7 mg/l) during 1983, with fairly high values during the whole ice-free season, compared to the very limited peaks at the beginning of the study period (Fig. 4 c).

Lake Vibollsjön has not been treated with lime, but it was affected by run-off water from Lake Iglafallssjön and the pH increased somewhat during the period 1977—82 (Table 3). This lake was devoid of fish and had a dense population of *Chaoborus*, with *C. flavicans* as the dominating species. *Cyclops* spp. and nauplii occurred in extremely low numbers in 1977, before liming of the upstream lake, but increased several-fold during the following year, at the same time as *Eudiaptomus gracilis* started to appear (Fig. 5). According to the net-hauls the population of *Chaoborus* increased markedly after 1977 (Fig. 5). During the whole study period, cladocerans were never found in the quantative samples, but single individuals of *Bosmina* sp., *Diaphanosoma brachyurum* and *Ceriodaphnia quadrangula* were occasionally found in net-hauls.

In Lake Svartsjön the abundance of zooplankton was rather low in 1976 and the major species were *Holopedium gibberum, Diaphanosoma brachyurum* and *Bosmina* sp. (Fig. 6 b). *Cyclops* spp. and nauplii were found in extremely low numbers (Fig. 6 a). In 1980, cladocerans were, however,

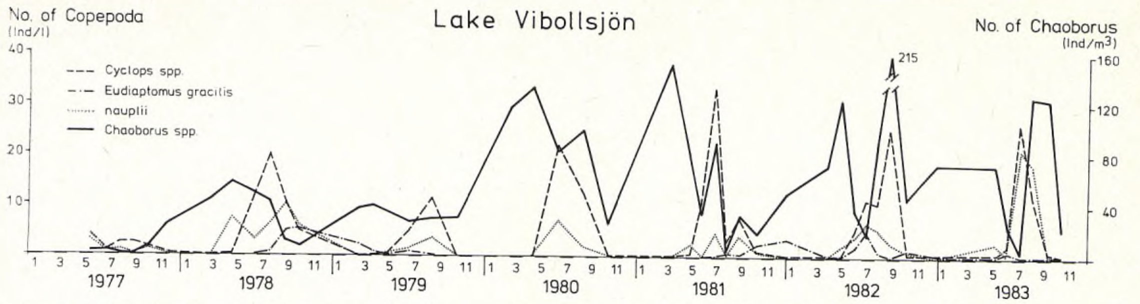


Fig. 5. Abundance of Copepoda and *Chaoborus* in Lake Vibollsjön 1977—83. (Volume-weighted averages for the water layer 0—7 m.)

totally absent and the very small crustacean community was made up of *Cyclops* spp. and nauplii. *Chaoborus flavicans* was not found in 1976 but frequently occurred in net-hauls in 1980 (Fig. 6 a).

In Lake Långsjön, the very sparse zooplankton community was made up of a few individuals of *Cyclops* spp., *Ceriodaphnia quadrangula* and *Bosmina* sp. during short periods in 1976. In 1980 *Ceriodaphnia quadrangula* and a very small number of *Bosmina* sp. were the only species found. The occurrence of these species in the samples was, however, of short duration (Fig. 7 b). *Chaoborus flavicans* was numerous in net-hauls in both years and seemed to have increased during 1980 (Fig. 7 a).

V. DISCUSSION

There are many differing opinions in the literature about the feeding selectivity of *Chaoborus* species. Older individuals, *i.e.* instars 3 and 4, seem, however, to feed mainly on crustaceans (Kajak and Ranke-Rybicka 1970, Swüste *et al.* 1973, Fedorenko 1975, Lewis 1977, Kajak and Rybak 1979, Lynch 1979, Smyly 1980, Swift and Forward 1981). Only Chimney *et al.* (1981) have found rotifers to be of significant importance as food for chaoborids. There are also different opinions about the crustacean species prefered by *Chaoborus*. Kajak and Ranke-Rybicka (1970), Swüste *et al.* (1973) and Swift and Forward (1981) found a preference for copepods in dif-

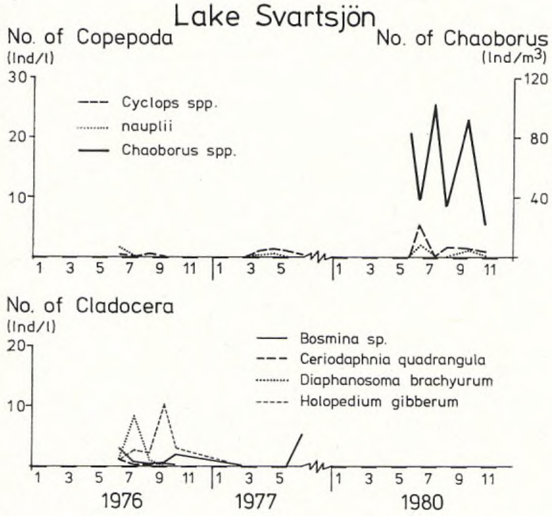


Fig. 6. Abundance of (a) Copepoda and *Chaoborus* and (b) Cladocera in Lake Svartsjön 1976—77 and 1980. (Volume-weighted averages for the water layer 0—7 m.)

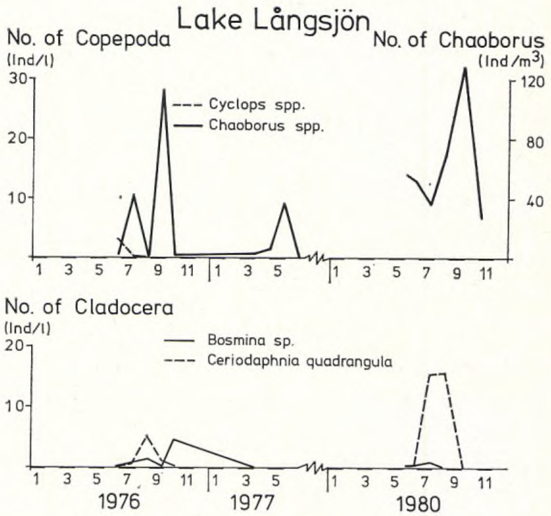


Fig. 7. Abundance of (a) Copepoda and *Chaoborus* and (b) Cladocera in Lake Långsjön 1976—77 and 1980. (Volume-weighted averages for the water layer 0—7 m.)

Table 4. *Average abundance of Copepoda, Cladocera (ind/l) and instars 3 and 4 of Chaoborus (ind/m³) in pelagic samples during May—October in Lakes Blanksjön, Iglafallssjön and Vibollsjön and June—October in Lakes Svartsjön and Långsjön.*

	1976	1977	1978	1979	1980	1981	1982	1983
L. Blanksjön								
Copepoda	—	3.9	11.2	14.2	18.9	5.7	8.7	11.9
Cladocera	—	0.3	1.3	1.4	17.8	11.3	11.3	20.4
Chaoborus	—	9.1	63.3	15.9	1.4	0	0.3	0.9
L. Iglafallssjön								
Copepoda	—	2.5	9.7	11.3	15.7	9.8	7.7	10.2
Cladocera	—	0.1	1.6	1.4	3.3	5.8	11.3	16.1
Chaoborus	—	32.7	39.2	37.4	10.9	0.2	0.6	1.1
L. Vibollsjön								
Copepoda	—	4.1	21.4	7.7	15.0	12.2	14.1	15.1
Cladocera	—	0	0	0	0.1	0	0.1	0.2
Chaoborus	—	3.1	28.0	29.3	70.0	43.6	67.1	62.2
L. Svartsjön								
Copepoda	0.6	—	—	—	2.4	—	—	—
Cladocera	8.0	—	—	—	0.2	—	—	—
Chaoborus	0	—	—	—	69.9	—	—	—
L. Långsjön								
Copepoda	0.6	—	—	—	0.2	—	—	—
Cladocera	2.7	—	—	—	5.6	—	—	—
Chaoborus	31.3	—	—	—	68.6	—	—	—

ferent stages of development. On the other hand, LEWIS (1977), LYNCH (1979) and SMYLY (1980) are of the opinion that different species of Cladocera, *e.g. Bosmina, Ceriodaphnia* and *Diaphanosoma* are easy to catch and are thus selected for, and WINNER and GREBER (1980) found a very strong selection for *Diaphanosoma.* KAJAK and RYBAK (1979) found no stable and distinct preference for any certain species and according to FEDORENKO (1975), the distributions of predator and prey were of primary importance for the food habits of *C. americanus* and *C. trivittatus.*

After the liming of Lakes Blanksjön and Iglafallssjön, the phytoplankton biomass decreased slightly in 1978. In 1979 it had approximately doubled compared to pre-liming levels in these lakes and in the downstream Lake Vibollsjön (ERIKSSON *et al.* 1982). There was also a shift from a dominance by Dinophyceae to a dominance by diatoms and chrysomonads in Lakes Blanksjön and Iglafallssjön (ERIKSSON *et al.* 1983). These changes in the phytoplankton ought to have favoured an increase in the biomass of zooplankton species. This did, however, not occur until 1980 in Lake Blanksjön and 1981 in Lake Iglafallssjön. In Lake Vibollsjön the composition of the crustacean community was still the same in 1983 as it was before the liming of the upstream lakes (Figs. 2, 4, 5).

There seems, however, to be a clear correlation between the number of cladoceran species and their density, and the abundance of chaoborids. In lakes with dense populations of chaoborids, such as Lakes Blanksjön 1977—79 (Fig. 2), Iglafallssjön 1977—80 (Fig. 4), Vibollsjön (Fig. 5), Svartsjön 1980 (Fig. 6) and Långsjön (Fig. 7), cladocerans were almost extinct (Table 4). This was not due to bad water quality, as all of the previously mentioned cladocerans except *Daphnia* occurred regularly in extremely small number in net-hauls. In Lake Svartsjön the water quality did not change between 1976 and 1980 (Table 3) and nevertheless cladocerans were almost extinct by the time that *Chaoborus flavicans* had become abundant in 1980. Moreover, the development of the cladoceran communities in Lakes Blanksjön and Iglafallssjön also coincided very well with the drastic decline of the chaoborid populations in 1979—80 and 1980—81, respectively (Figs. 2, 4, Table 4).

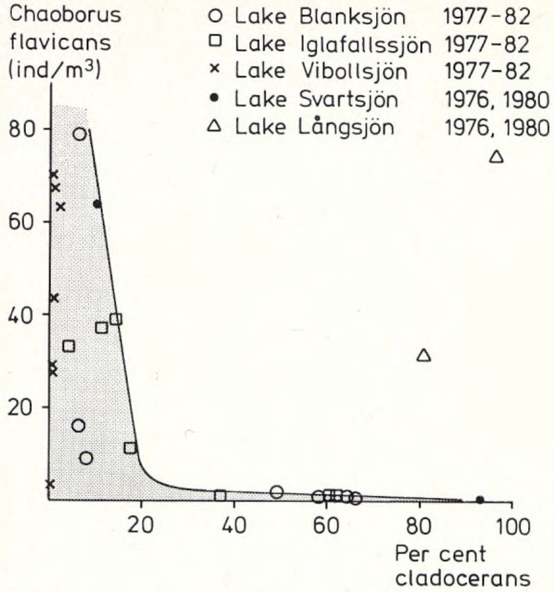


Fig. 8. Abundance of *Chaoborus flavicans* and proportion of cladocerans, in per cent of the total number of crustaceans, in the lakes studied. (Volume-weighted averages calculated for 5—6 occasions during May—October.)

When looking at the average abundance of *Chaoborus* during May—October in pelagic samples, one finds that cladocerans are the dominating crustaceans when chaoborid density is low (Table 4), and that the proportion of cladocerans as a percentage of the total number of planktonic crustaceans decreased with increasing abundances of *Chaoborus* in the lakes studied (Fig. 8). The figure has little general validity as the migratory behaviour of *Chaoborus* varies with instar stage and also with the seasons (GOLDSPINK and SCOTT 1971). Moreover, only one single net-haul was taken on each sampling occasion. Nevertheless, it seems that a *Chaoborus* abundance of less than 10 ind/m^3 in pelagic samples is enough to reduce the cladocerans in these lakes. At *Chaoborus* densities greater than 10 ind/m^3, cladocerans constituted less than 20 per cent of the total number of planktonic crustaceans. The results from Lake Långsjön are, however, quite different as cladocerans dominated the very weak community in both the years studied (Fig. 8).

The apparent preference of *Chaoborus* for cladocerans probably depends on the fact that cladocerans are easier to catch than copepods. It has also been shown that *Mysis relicta* (LASENBY and FÜRST 1981), whitefish larvae (*Coregonus*) (BRAUM 1967) and most fish species (NILSSON and PEJLER 1973) have difficulties in catching copepods, possibly because of their efficient escape behaviour.

In order to try and get an idea of the magnitude of the impact of the predation by *C. flavicans* on the crustacean communities in Lakes Blanksjön and Iglafallssjön, the consumption of food by instars 3 and 4 of the predator was calculated according to KAJAK and RYBAK (1979). The calculated consumption was fairly low (0.07—0.20 mg ww/l · month) in Lake Blanksjön during July—September 1977 (Table 5), but increased several-fold to 0.15—1.14 mg/l during June—September 1978, due to an increase in the biomass of *C. flavicans* after liming. Predation by brown trout reduced the biomass of *C. flavicans* in late summer 1979 and the food consumption of the chaoborid population decreased.

In Lake Iglafallssjön, the food consumption of *C. flavicans* was high during 1977, when the population was dense, and there was only a slight increase in their consumption during 1978. During June—September 1980 the chaoborid population was sparse and food consumption was very low (Table 5).

If *C. flavicans* had fed exclusively on crustaceans, ca 0.5—2.5, 0.6—14 and 0.2—5.8 times the biomass would have been consumed monthly in Lake Blanksjön during the summers of 1977, 1978 and 1979 respectively. In Lake Iglafallssjön ca 4.1—10.2, 0.8—7.0, 0.6—3.8 and 0.1—0.5 times the crustacean biomass would have been consumed monthly during the same years and in 1980 (Table 5).

This calculation may have been biased in a number of ways. One of the critical values is the assumed hourly ration of 0.5 % of the chaoborids' fresh weight. It is questionable whether *C. flavicans* feeds continously during the active period, the length of which has also been assumed according to KAJAK and RYBAK (1979). Further, the calculated chaoborid biomass is somewhat too high for June 1978 and 1979 in Lakes Blanksjön and Iglafallssjön, as the population estimates were

Table 5. *Calculated consumption of crustaceans by instars 3 and 4 of Chaoborus flavicans.*

Date	Number of Chaob. (ind/m³)	Mean weight of Chaob. (mg/ind)	Biomass of Chaob. (mg/l)	Feeding hours per day	Calculated consumption (mg/l · month)	Calculated biomass of crustaceans (mg/l)	% crustacean biomass consumed monthly by Chaob.
L. Blanksjön							
1977 July	37.6	1.4	0.053	8.0	0.07	0.13	54
Aug.	,,	2.3	0.086	14.0	0.19	0.15	127
Sept.	,,	2.8	0.105	12.5	0.20	0.08	250
1978 June	,,	4.9	0.184	5.5	0.15	0.08	188
July	218.0	1.4	0.305	8.0	0.38	0.60	63
Aug.	,,	2.3	0.501	14.0	1.05	0.65	162
Sept.	,,	2.8	0.610	12.5	1.14	0.08	1425
1979 June	128.2	4.9	0.628	5.5	0.52	0.09	578
July	33.7	1.4	0.047	8.0	0.06	0.27	22
Aug.	,,	2.3	0.078	14.0	0.16	0.33	49
Sept.	,,	2.8	0.094	12.5	0.18	0.53	34
L. Iglafallssjön							
1977 July	102.9	1.4	0.144	8.0	0.18	0.03	600
Aug.	,,	2.3	0.237	14.0	0.51	0.05	1020
Sept.	,,	2.8	0.288	12.5	0.54	0.13	415
1978 June	,,	4.9	0.504	5.5	0.42	0.11	382
July	120.1	1.4	0.168	8.0	0.21	0.28	75
Aug.	,,	2.3	0.276	14.0	0.58	0.30	193
Sept.	,,	2.8	0.336	12.5	0.63	0.09	700
1979 June	42.5	4.9	0.208	5.5	0.17	0.08	213
July	85.7	1.4	0.120	8.0	0.15	0.27	56
Aug.	,,	2.3	0.197	14.0	0.41	0.23	178
Sept.	,,	2.8	0.240	12.5	0.45	0.12	375
1980 June	76.7	4.9	0.376	5.5	0.31	0.57	54
July	9.4	1.4	0.013	8.0	0.02	0.38	5
Aug.	,,	2.3	0.022	14.0	0.05	0.15	33
Sept.	,,	2.8	0.026	12.5	0.05	0.27	19

made in April. On the other hand, the calculated biomass and hence the consumption is too low during July—September every year, as the mortality between the period July—September and the sampling occasion in December has not been taken into consideration.

The calculated crustacean biomass seems to be in good agreement with results from other oligotrophic lakes in central Sweden. JOHANSSON *et al.* (1976) found a maximum average biomass of ca. 280 mg dw/m² and an average summer biomass of ca. 200 mg dw/m² of filter-feeding crustaceans during the years 1972—75 in Lake Botjärn. These figures correspond to 0.85 mg ww/l and 0.61 mg ww/l respectively if one assumes the dry weight to be 10 % of the fresh weight (BOTTRELL *et al.* 1976). According to PERSSON (1977) the summer biomass is in the range of 0.5—1.0 mg ww/l in oligotrophic lakes in different parts of Sweden.

If the calculations are reasonable, they imply that the population of *C. flavicans* cannot feed exclusively on crustaceans. In this case the calculated consumption corresponds to monthly maximum P/B-ratios of 2.5, 14.3 and 5.8 and 10.2, 7.0 and 3.8 for the crustacean communities in Lakes Blanksjön and Iglafallssjön respectively, during 1977—79. According to MORGAN *et al.* (1980), the monthly P/B-ratios of herbivorous crustaceans should be around 3. The conclusion is that *C. flavicans*, after having drastically reduced the crustacean populations, has to feed on other food items, *e.g.* rotifers. It is thus possible that the divergent results found in the literature concerning the food preference of *Chaoborus* in natural waters may, at least in lakes devoid of fish, be explained by the magnitude of the predation by *Chaoborus*. CHIMNEY *et al.* (1981) for example, found rotifers to be abundant prey items in a lake devoid of cladocerans and with a very sparse population of copepods. The structure of this zooplankton community may very well have been an effect of intensive predation by chaoborids.

The magnitude of the impact of chaoborid predation on zooplankton communities will of course be a function of the abundance of these predators. As *Chaoborus* species are highly susceptible to predation by fish (NORTHCOTE *et al.* 1978, STENSON 1978), the impact of chaoborids will be most pronounced in waters devoid of fish. Nevertheless, NEILL (1981) found that chaoborids had a negative effect on the development of *Bosmina* and *Diaphanosoma* in particular, during periods of low reproduction for the two latter species. KAJAK and RYBAK (1979) also found that predation by *Chaoborus* was of great significance and that in August it even exceeded crustacean production in the central zone of Lake Mikolajskie.

After liming of the fish-free Lake Gårdsjön, HENRIKSON *et al.* (1984) found a decreased abundance of *Bosmina* spp. and an increased abundance of *Diaphanosoma*. They conclude that this is due to increased predation on *Bosmina* by *Chaoborus* and the effect on *Diaphanosoma* of decreased competition for food with *Bosmina*. The effects of *Chaoborus* predation are smaller than in the present study, which may be attributed to the fact that the abundance of chaoborids was less than 250 ind/m^2 (HENRIKSON and OSCARSON 1984), compared to maximum values of ca 1000 and 670 ind/m^2 in Lakes Blanksjön and Iglafallssjön respectively.

The results from the lakes in this study are almost in exact agreement with the results of LYNCH (1979), with the sole exception that *Daphnia* species are missing during acid conditions. LYNCH (1979) also stated that in the absence of fish, *Chaoborus* was able to eradicate herbivorous crustaceans.

VI. REFERENCES

ALMER, B., W. DICKSON., C. EKSTRÖM and E. HÖRNSTRÖM, 1978. Sulfur pollution and the aquatic ecosystem. p. 272—311. *In* Sulfur in the Environment: 11. Ecological Impact. Ed: J. O. Nriago. John Wiley & Sons, New York.

BOTTRELL, H. H., A. DUNCAN, Z. M. GLIWICZ, E. GRYGIEREK, A. HERZIG, A. HILLBRICHT-ILKOWSKA, H. KURASAWA, P. LARSSON and T. WEGLEŃSKA. 1976. A revue of some problems in zooplankton production studies. Norw. J. Zool. 24: 419—456.

BRAUM, E., 1967. The survival of fish larvae with reference to their feeding behaviour and the food supply. p. 113—131. *In* The biological basis of freshwater fish production. Ed.: S. D. Gerking. Blackwell Scientific Publications.

BROOKS, J. L. and S. J. DODSON. 1965. Predation, body-size and composition of plankton. Science 158: 28—35.

CHIMNEY, M. J., R. W. WINNER and S. K. SEILKOP. 1981. Prey utilization by *Chaoborus punctipennis* SAY in a small, eutrophic reservoir. Hydrobiologia 85: 193—199.

ERIKSSON, F., E. HÖRNSTRÖM, P. MOSSBERG and P. NYBERG. 1982. Ecological effects of lime treatment of acidified lakes and rivers. Inf. Inst. Freshw. Res., Drottningholm (6). 96 p. (In Swedish with English summary.)

— E. HÖRNSTRÖM, P. MOSSBERG and P. NYBERG. 1983. Ecological effects of lime treatment of acidified lakes and rivers in Sweden. Hydrobiologia 101: 145—164.

ERIKSSON, M. O. G., L. HENRIKSSON, B.-I. NILSSON, G. NYMAN, H. G. OSCARSON, A. E. STENSON and K. LARSSON. 1980. Predatory-prey relations important for the biotic changes in acidified lakes. Ambio 9: 248—249.

FEDORENKO, A. Y., 1975. Instars and species-specific diets in two species of Chaoborus. Limnol. Oceanogr. 20: 238—249.

GALBRAITH JR., M. G. 1967. Size-selective predation on Daphnia by rainbow trout and yellow perch. Trans. Am. Fish. Soc. 96: 1—10.

GLIWICZ, Z. M., A. HILLBRICHT-ILKOWSKA and T. WEGLEŃSKA. 1978. Contribution of fish and invertebrate predation to the elimination of zooplankton biomass in two Polish lakes. Verh. Internat. Verein. Limnol. 20: 1007—1011.

GOLDSPINK, C. R. and D. B. C. SCOTT. 1971. Vertical migration of *Chaoborus flavicans* in a Scottish loch. Freshw. Biol. 1: 411—421.

GRÖNBERG, B. 1973. Investigations of zooplankton in Ekoln 1967—1969. Medd. NLU 54. 36 p. (In Swedish with English abstract.)

HARVEY, H. H. 1980. Widespread and diverse changes in the biota of North American lakes and rivers coincident with acidification. p. 93—98: *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

HENRIKSON, L. and H. OSCARSON. 1984. Lime influence on macroinvertebrate zooplankton predators. Rep. Inst. Freshw. Res., Drottningholm 61: 93—103.

— H. OSCARSON and J. STENSON. 1984. Development of the crustacean zooplankton community after lime treatment of the fishless Lake Gårdsjön, Sweden. Rep. Inst. Freshw. Res., Drottningholm 61: 104—114.

Hobaek, A. and G. G. Raddum. 1980. Zooplankton communities in acidified lakes in South Norway. SNSF-project IR 75/80. Oslo—Ås. 132 p.

Johansson, J. Å., H. Olofsson and L. Ramberg. 1976. Studier av zooplanktons konsumtion i Botjärn. Klotenprojektet Rapp. 5. Inst. Limnol. Univ. Uppsala. (Mimeographed in Swedish.) 27 p.

Johansson, K. and P. Nyberg. 1981. Acidification of surface waters in Sweden — effects and extent 1980. Inform. Inst. Freshw. Res., Drottningholm 6. 118 p. (In Swedish with English summary.)

Kajak, Z. and B. Ranke-Rybicka. 1970. Feeding and production efficiency of *Chaoborus flavicans* Meigen (Diptera, Culicidae) larvae in eutrophic and dystrophic lake. Pol. Arch. Hydrobiol. 17: 225—232.

— and J. Rybak. 1979. The feeding of *Chaoborus flavicans* Meigen (Diptera, Chaoboridae) and its predation on lake zooplankton. Int. Rev. Hydrobiol. 64: 361—378.

Lasenby, D. C. and M. Fürst. 1981. Feeding of *Mysis relicta* Lovén on macrozooplankton. Rep. Inst. Freshw., Drottningholm. 59: 75—80.

Lewis, W. M. 1977. Feeding selectivity of a tropical *Chaoborus* population. Freshw. Biol. 7: 311—325.

Lynch, M. 1979. Predation, competition and zooplankton community structure: An experimental study. Limnol. Oceanogr. 24: 253—272.

Morgan, N. C., T. Backiel, G. Bretschko, A. Duncan, A. Hillbricht-Ilkowska, Z. Kajak, J. F. Kitchell, P. Larsson, C. Levêque, A. Nauwerck, F. Schiemer and J. E. Thorpe. 1980. Secondary production. p. 247—340. *In* The functioning of freshwater ecosystems. Eds: E. D. Le Cren and R. H. Lowe-Mc Connell. International Biological Programme 22, Cambridge University Press.

National Swedish Environment Protection Board. 1981. Kodlista 80. Vattenundersökningar. SNV PM 1492. 277 p. (In Swedish.)

Neill, W. E. 1981. Impact of *Chaoborus* predation upon the structure and dynamics of a crustacean zooplankton community. Oecologia 48: 164—177.

Nilsson, N.-A. and B. Pejler. 1973. On the relation between fish fauna and zooplankton composition in North Swedish lakes. Rep. Inst. Freshw. Res., Drottningholm 53: 51—77.

Northcote, T. G., C. J. Walters and J. M. B. Hume. 1978. Initial impact of experimental fish introductions on the macrozooplankton of small oligotrophic lakes. Verh. Internat. Verein. Limnol. 20: 2003—2012.

Persson, G. 1977. Zooplankton 1971—1976. p. 141—160. *In* Kuokkel Progress. Rep. 5. Inst. Limnol., Univ. Uppsala. (In Swedish with English summary.)

Schofield, C. L. 1976. Acid precipitation: Effects on fish. Ambio 5: 228—230.

Sevalrud, I. H., I. P. Muniz and S. Kalvenes. 1980. Loss of fish populations in southern Norway. Dynamics and magnitude of the problem. p. 350—351. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

Smyly, W. J. P. 1980. Food and feeding of aquatic larvae of the midge *Chaoborus flavicans* (Meigen) (Diptera: Chaoboridae) in the laboratory. Hydrobiologia 70: 179—188.

Stenson, J. A. E. 1978. Differential predation by fish on two species of *Chaoborus* (Diptera, Chaoboridae). Oikos 31: 98—101.

— T. Bohlin, L. Henrikson, B. J. Nilsson, H. G. Nyman, H. G. Oscarsson and P. Larsson. 1978. Effects of fish removal from a small lake. Verh. Internat. Verein. Limnol. 20: 794—801.

Swift, M. C. and R. B. Forward Jr. 1981. *Chaoborus* prey capture efficiency in the light and dark. Limnol. Oceanogr. 26: 461—466.

Swüste, H. F. J., R. Cremer and S. Parma. 1973. Selective predation by larvae of *Chaoborus flavicans* (Diptera, Chaoboridae). Verh. Internat. Verein. Limnol. 18: 1559—1563.

Winner, R. W. and J. S. Greber. 1980. Prey selection by *Chaoborus punctipennis* under laboratory conditions. Hydrobiologia 68: 231—233.

Effects of Lime Treatment on The Benthos of Lake Søndre Boksjø

GUNNAR G. RADDUM,[1] GØSTA HAGENLUND [2] and GODTFRED A. HALVORSEN [1]

[1] Museum of Zoology, University of Bergen, Muséplass 3, N-5000 Bergen, Norway.
[2] Zoological Institute, University of Oslo, P. O. Box 1051, Blindern, N-0316 Oslo 3, Norway.

ABSTRACT

Lake Søndre (=S.) Boksjø, on the border between Sweden and Norway, was during July-August 1980 limed with 9000 tons of $CaCO_3$.

Quantitative and qualitative samples of bottom animals were taken before and after the treatment in Lake S. Boksjø and in the untreated lake, Nordre (=N.) Boksjø. The running water fauna was investigated at one locality downstream of Lake N. Boksjø and at two localities downstream of Lake S. Boksjø.

The mayfly *Baetis rhodani* (PICTET) was absent from the river before liming. After the treatment the species occurred at the upper part of the outlet river of Lake S. Boksjø. *Asellus aquaticus* (L.) was present in slow-flowing parts of the river before liming. After the treatment the species increased in density 8—10 times.

A. aquaticus was not found in Lake S. Boksjø before liming, but occurred with a few individuals after the treatment.

Increased abundance was recorded in the treated lake for the mayflies *Leptophlebia* spp., oligochaetes and chironomids in the depth zone 0.5—5 m. This was probably due to increased food availability for detritivorous animals, caused by the liming.

The increased number of species of coleopterans and corixids observed was probably caused by elevated biological activity in general.

In the profundal of Lake S. Boksjø a strong decrease in number of species and density of bottom animals was recorded after liming. This is suggested to be caused by precipitation of Al or other metals.

I. INTRODUCTION

In Scandinavia, USA and Canada, acidification of lakes and rivers has been one of the most serious environmental problems (SCHOFIELD 1976, ALMER *et al.* 1978, DRABLØS and TOLLAN 1980, SWEDISH MINISTRY OF AGRICULTURE 1982). Liming of lakes and rivers in Scandinavia to counteract the acidification of valuable waterbodies, has been carried out for several years. (ERIKSSON *et al.* 1983, KALKINGSPROSJEKTET 1983).

In 1980 the upper part of the Boksjø/Kornsjø watershed on the border of Sweden and Norway was limed, starting with Lake S. Boksjø.

On behalf of the Norwegian liming project the Universities of Bergen and Oslo carried out investigations on the bottom fauna in the watershed from 1980 to 1983. Parts of this investigations will be published later.

The investigations of water chemistry, phytoplankton, zooplankton and fish were carried out by Swedish scientists.

Positive effects of lime treatment of acidic lakes on phytoplankton, zooplankton and benthos have been reported by HENRIKSON *et al.* (1981), HULTBERG and ANDERSSON (1981) and ERIKSSON *et al.* (1983) but there still remains a great need for information, especially about effects on the benthos.

II. THE STUDY AREA

The investigated part of the watershed is shown in Fig. 1. The Lake N. Boksjø was not limed and served as a control lake. The Swedish part of Lake S. Boksjø (east side) was limed during July—August 1980 with 9000 tons of $CaCo_3$, mostly at the shoreline.

It was estimated that this amount of lime should elevate the pH of the lake water from about 4.5

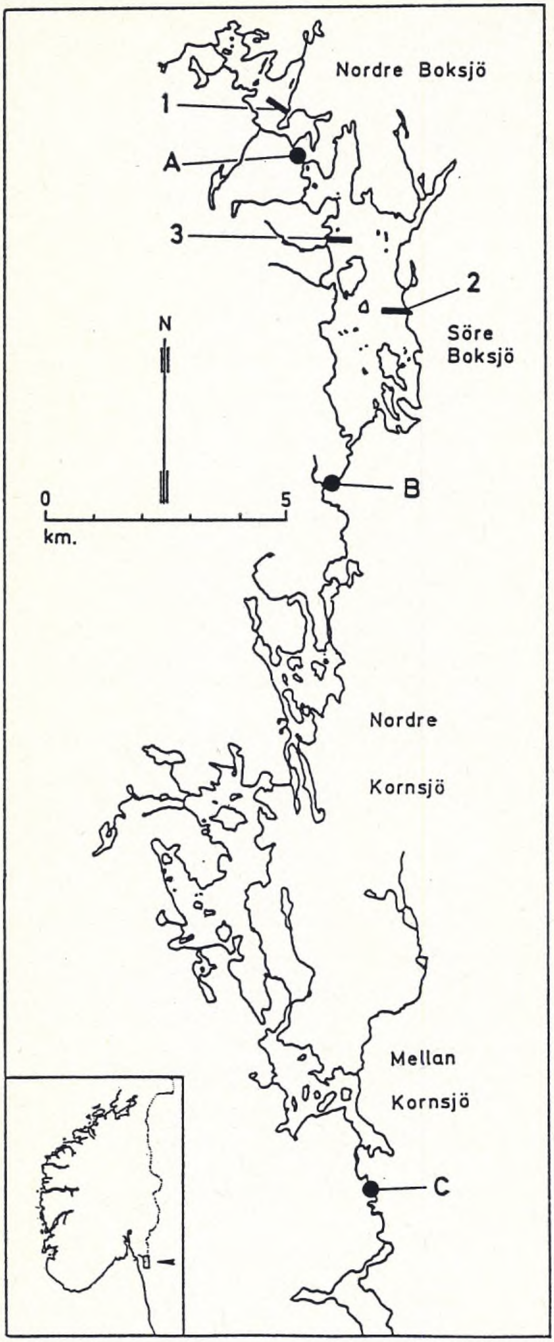


Fig. 1. Map of the investigated watershed. Localities in lakes marked with numbers and in running water with letters.

to 6.5—7.0 and compensate the acidification for the next 5 years.

Some physical and chemical data of the lakes are given in Tables 1 and 2.

Before liming both lakes were acidic clearwater lakes with pH of 4.5—4.8. However, Lake N. Boksjø was regarded as the most humic one. Ca+Mg content was low (2.4—3.4 mg/l), while total Al reached high values (about 250 µg/l), typical of acidified lakes.

After liming the pH increased to 6.7—7.0 and alkalinity to 0.11—0.13. The Ca+Mg hardness was therefore doubled, while the total Al decreased to a range of 54—137 µg/L (Table 2).

Effects of the treatment were seen downstream in a reducing trend from Lake N. Kornsjø to Lake S. Kornsjø. In Lake N. Kornsjø, which is representative for our locality C (Fig. 1) the pH increased from the range 4.8—5.3 to > 6. Also alkalinity and hardness of the water have increased (DICKSON pers. comm.).

The sampling locality in Lake N. Boksjø (station 1, Fig. 1) has a thick layer of dy typical for lakes in coniferous regions of Scandinavia. The littoral zone consists of rocks with dy bottom in between, and some vegetation of eledoids and isoëtids.

Lake S. Boksjø was more wind-exposed than Lake N. Boksjø. The bottom was irregular with exposed rocks down to at least 25 m. Between the barren rocks, deposits of dy or gyttja existed. At station 3 the bottom was more flattened out with a thicker layer of dy. In the littoral zone, rocks and sand/gravel dominated, but in wind-protected bays silt bottoms existed with vegetation of eledoids and isoëtids.

Station A, B and C were running water localities, representing unlimed (A) and limed situations (B and C).

The water current was rapid at A and B and slow at C which had a dense zone of macrovegetation along the shore.

III. METHODS

Qualitative samples

Substrate both from the littoral zone and running water zone was sampled by a kicking method

Table 1. *Physical characteristics of the investigated lakes. (Data taken from* Borgstrøm *et al. 1974,* Björn Lindquist, *Älvsborgs Läns Norra Fiskevårdsförbund and* G. Hagenlund *unpubl.).*

	Catchment area (ha)	Surface area (ha)	Max. depth	Mean depth	Volume (mil. m^3)	Colour	Mean transparency (m)
Lake N. Boksjø	—	210	18	8.3	10	yellow	5.4
Lake S. Boksjø	5200	810	57	14.0	13	green	11.8

(Frost *et al.* 1971, Brittain and Lillehammer 1978). The sampled substrate was preserved in ethanol and sorted in the laboratory. Three to five samples were taken at each station depending on the type of substrate. Area and sampling time was kept constant for each station in order to make comparisons between the localities possible.

Quantitative samples

The quantitative measurements were carried out by use of a core sampler. At each depth (0.5, 2, 5, 10 m and the deepest part) 5—7 samples were taken. However, due to hard bottom, especially at station 2, not all depths were always sampled. The substrate was sieved through a 0.25 mm net and sorted under microscope in the laboratory.

IV. RESULTS AND DISCUSSION

The composition of the animal communities in the investigated localities was of a typical acidic character before liming. Sensitive organisms like gastropods were not recorded, whereas bivalves were found, one specimen from Lakes N. and S. Boksjø, respectively. Oligochaetes were recorded in low numbers in the littoral zone. Below 5 m depths these animals were usually not found.

The mayflies (Ephemeroptera) in both lakes consisted of *Leptophlebia* spp. At locality A and B they were absent but at C *Leptophlebia* spp., *Siphlonurus linneanus* (Eaton) and *Heptagenia* sp. were recorded before the treatment.

The stoneflies (Plecoptera) and caddisflies (Trichoptera) were represented with species typical for acidic water (Raddum and Fjellheim 1984). The species regarded as sensitive to acid water were not recorded neither before nor after liming. The above mentioned groups will not be discussed here, as they will appear in a later paper.

The qualitative measurements, mean number of animals caught per min. before and after liming, is shown in Fig. 2.

Both in 1980 and 1981 the total number of animals recorded was low in Lake N. Boksjø. In Lake S. Boksjø the numbers were higher before liming. After liming the number of specimens increased about 3-fold in spring and autumn at locality 3 and 2, respectively. This was mainly

Table 2. *Chemical characteristics of Lakes N. Boksjø and S. Boksjø, surface water, during the period 1979—83. (From* Swedish Ministry of Agriculture *(1982) and* W. Dickson *pers. comm.).*

	pH	Alk.	Ca+Mg mg/l	Al µg/l
Lake N. Boksjø (Range during the period 1979—83)	4.5—4.8	0	2.4—2.6	215—258
Lake S. Boksjø (Range during the period 1979 to liming)	4.5—4.8	0	2.6—3.2	250—279
Lake S. Boksjø (Range after liming to May 1983)	6.7—7.0	0.1—0.13	5.6—6.2	54—137

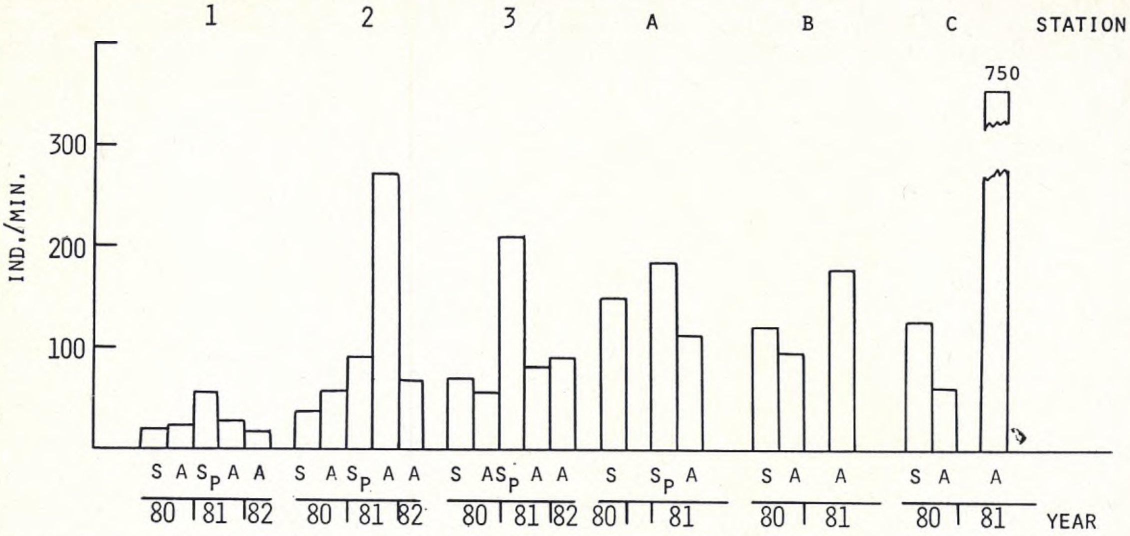


Fig. 2. Mean number of animals caugth per min at different time and localities.
S=summer, A=autumn, Sp=spring.

caused by the occurrence of a high fraction of coleopterans (Table 3).

Phyto- and zooplankton populations increase rapidly after liming and *Sphagnum* spp. decrease considerably (Eriksson *et al.* 1983). This effect would increase food resources to most of the coleopterans.

On the species level, the coleopterans were dominated by *Coelambus novemlineatus* (Steph.), and *Gyrinus aeratus* Steph. both before and after liming (Table 3). Further, the species *Platambus maculatus* (L.) and *Agabus sturmi* (Gyll.) appeared only after the treatment.

All species select larger bodies of water for reproduction and were considered physiologically insensitive to the pH range.

The predation of fish on coleopterans in Lake S. Boksjø was low in 1980 (Thörnelöf 1982). The situation in 1981 was probably much the same allowing the species to expand.

Coleopterans are considered to be of great importance as fish food in recently limed acidic lakes (Sveälv and Hagenlund in prep.), due to high caloric content (Driver 1981) and high abundance. However, they are in general very susceptible to fish predation (Stenson 1979) and most species will probably be reduced in numbers with increasing fish population.

Asellus aquaticus (L.), an important prey species for fish, was not found in the lakes before liming. After the treatment, the species was found at locality 2.

Corixids (Hemiptera) were represented in the lakes by a few species (Table 3). This taxon was heavily predated in 1980 (Thörnelöf 1982). *Cymatia bonsdorffi* (Sahlb.) was quite frequent before and after the treatment, while *Glaenocorisa propinqua* (Fieb.) and *Hesperocorixa castanea* (Thoms.) were found only after. It was not possible to make any conclusions about these communities in the lakes, caused by liming.

In the running water localities A and B, no marked differences in total animal densities were observed to occur during the research periode. However, at locality C, a very distinct increase in animal abundance, about 7 fold, was noted in the autumn of 1981.

On the species level some changes occurred after liming: The mayfly *Baetis rhodani* (Pictet) was not found before liming neither at locality A nor B, which are typical habitats for this species. After the lime treatment the larvae of *B. rhodani* became relatively abundant at B in the autumn 1981).

In western Norway this species is regarded as an important indicator of less acidic running water

Table 3. *Relative occurrence of different species of Coleoptera, Hemiptera and Odonata.* ××× — *dominating,* ×× — *common,* × — *common in low number,* (×) — *rare.*

	Before liming				After liming			
	1	2	3	C	1	2	3	C
Coleoptera:								
Hyphydrus ovatus (L.)				×	×			(×)
Hygrotus quinquelineatus (ZETT.)	×	×	×		×	×	×	
Coelambus novemlineatus (STEPH.)	×	×××	×		×	×××	×××	
Hydroporus palustris (L.)		(×)					(×)	
H. erythrocephalus (L.)		(×)				×	×	
Graptodytes pictus (FABR.)		(×)			(×)	×		
Potamonectes depressus (FABR.)	×	×	×		×	×	×	
Platambus maculatus (L.)						×		
Agabus sturmi (GYLL.)							×	
Ilybius aenescens THOMS.	×	(×)	×					
Rhantus exsoletus (FORST.)	×		×	××	(×)		×	×
Hydroporus sp. larvae		×						
Agabus sp. larvae				(×)				(×)
Gyrinus aeratus STEPH.	××	×××	××	×	×	×××	×××	××
G. marinus GYLL.				×				
G. substriatus STEPH.				×				×
G. minutus FABR.				×				×
Gyrinus sp. larvae	×		×	××				
Galerucella sp.								(×)
Hemiptera:								
Cymatia bonsdorffi (SAHLB.)	××	×	×	×××			×	××
Glaenocorisa propinqua (FIEB.)							×	
Hesperocorixa linnei (FIEB.)								×
H. castanea (THOMS.)							×	
Sigara striata (L.)								××
S. semistriata (FIEB.)				×				×××
S. fossarum (LEACH)				×××				×××
Callicorixa praeusta (FIEB.)								×
C. wollastoni (DOUGLAS & SCOTT)								×
C. producta (REUT.)								×
Corixidae nymphs indet.		××		×××				
Notonecta glauca L.			×	×				
Gerris sp.				×				×
Megaloptera:								
Sialis lutaria (L.)		×	×					
Odonata:								
Coenagrion hastulatum CHARP.								××
Leucorrhynia pectoralis (CHARP.)					×	×	×	
[1]								

[1] *Cordulegaster boltoni* DONOWAN and *Somatochlora metallica* (LIND.) was recorded at station A and B after liming.

(RADDUM and FJELLHEIM 1984). Experiments carried out by RADDUM (1979) showed that the larvae died quickly in soft water with pH≈4.5—4.7, while a few larvae were able to survive this acidity in water with higher content of ions.

In Germany both field observations and experiments support the conclusion that *B. rhodani* avoid acidic water (MATTHIAS 1982). SUTCLIFFE and CARRICK (1973) observed that the larvae were absent from stretches of River Duddon with pH < 5.7 and that the imagoes do not drop their eggs in water of that acidity.

3,500 localities in Sweden have been investigated regarding mayflies and acidity (ENGBLOM and LINGDELL 1983). *B. rhodani* was most frequently found in water with pH 6.0—6.9, but larvae have

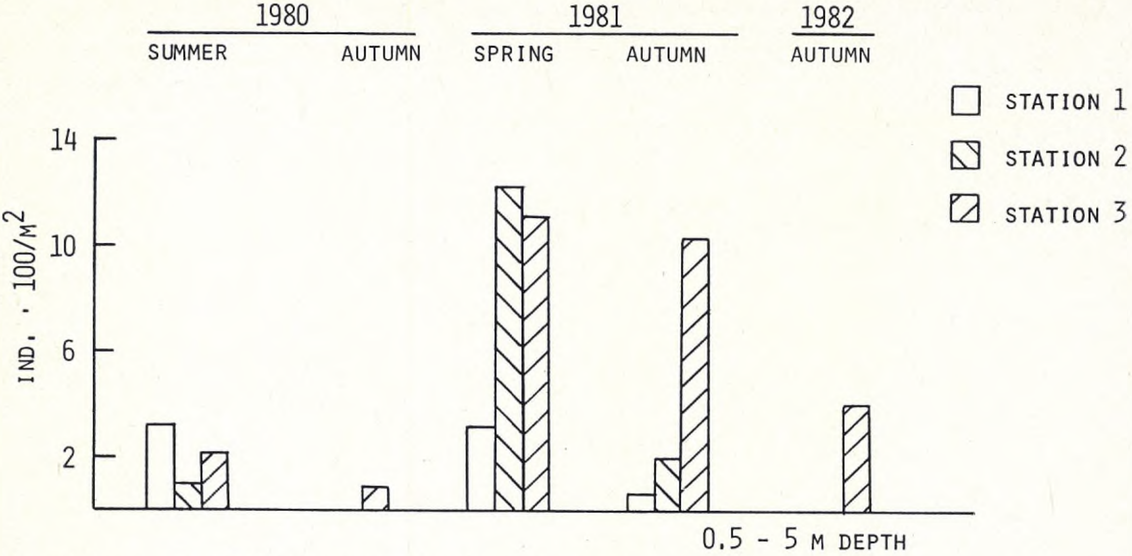


Fig. 3. Mean number of oligochaetes per m² at the depth-zone 0.5—5 m.

been observed in water with pH as low as 4.6. This observation was confirmed by experiments. However, no data about the conductivity and water quality was given. It is, however, suggested that precipitation of Al during increasing pH of the water, can be an important factor in wiping out *B. rhodani*.

After lime treatment of Lake S. Boksjø Al precipitated and the pH rose to optium values for *B. rhodani* (SWEDISH MINISTRY OF AGRICULTURE 1982). We suggest that the colonization of the species in the main river at B is a direct result of the liming and that *B. rhodani* probably has immigrated from refuges in the surroundings.

The high number of animals recorded at locality C consisted mainly of mayflies.

Leptophlebia vespertina (L.) and *L. marginata* (L.) dominated in 1980. In the autumn 1981 these species — together with *Heptagenia fuscogrisea* (RETZIUS) had developed very dense populations.

The above-mentioned species are widely spread in Sweden (ENGBLOM and LINGDELL 1983) and are able to survive in the most acidified watersheds. They are doubtless favoured by liming, during absence of other insects.

S. linneanus, which was found in low numbers before liming, was not recorded after.

A. aquaticus, a common species before the treatment, increased in density 8—10 fold after liming in the autumn 1981. The species was favoured by liming (ERIKSSON *et al.* 1983), but the response in our case is very dramatic and will probably be of short duration.

The corixid fauna also increased after liming at site C, both in density and number of species present. The highest positive effect was noted in *Sigara striata* (L.), *Callicorixa praeusta* (FIEB.), *C. wollastoni* (DOUGLAS and SCOTT), *C. producta* (REUT.) and *S. fossarum* (LEACH) populations. Except for the last named, these species were not found in 1980.

Due to the insects' yearly fluctuations it is difficult to draw a conclusion about the observed change in the corixid fauna at locality C. We believe, however, that the increase is actually a result of elevated biological activity after the treatment. Because locality C contained a very high amount of organic material there was a great increase of detritus-living animals. The availability of this material as food for invertebrates has probably increased due to the liming. The detritivorous animals seemed to react immediately on the unexploited resources, which will have a positive effect on the whole community.

The quantitative measurements in the lakes are shown for oligochaetes and chironomids in Figs. 3 and 4.

The highest abundance of oligochaetes was found in the littoral/sublittoral zone (0.5—5 m). At 5 m their occurrence was very irregular, while the animals were nearly absent in deeper parts. Before liming the mean number of oligochaetes in the depth zone 0.5—5 m was low at the three localities (< 300 ind. per m²).

However, an increase occurred both at locality 2 and 3 in spring 1981 (1100—1200 ind. per m²). The following autumn the abundance remained high at site 3, but was reduced at site 2. By 1982 the oligochaete density seemed to lie at the pre-limed density level once again.

The chironomids dominated the bottom fauna with the highest abundance in the littoral zone. The pre-limed sampling data showed low densities of chironomids at all depths, both in Lakes S. Boksjø and N. Boksjø (Fig. 4). After the treatment in spring 1981 an effect on the chironomid fauna was first observed at loc. 3 at 5 m depth. In autumn 1981 the liming effect had expanded to the 0.5—2 m depth zone. A year later, maximum values were measured at 14,355 — and 8,015 larvae per m² in the littoral — and 5 m depth zone, respectively. These values are 3—4 times higher than those measured at the control site (loc. 1).

At locality 2, however, only a limited response was noted at 5 m depth in the spring and autumn 1981 (Fig. 4).

It is possible that the amount of organic material is the determining factor for the positive response of liming on the chironomids at depths ≤ 5 m at locality 3.

Table 4 shows the species composition of chironomids in the two lakes in the autumn 1982. The composition is more or less similar to earlier reports from acidified waters (Wiederholm and Erikson 1977, Mossberg and Nyberg 1979, and Raddum and Sæther 1981).

The result after liming in Lake S. Boksjø, compared with the unlimed Lake N. Boksjø fits also well with the descriptions from other lime-treated lakes given by Eriksson *et al.* (1983).

Heterotanytarsus apicalis (Kieff.) which was recorded in very low number in the profundal in Lake N. Boksjø, was one of the dominating species in Lake S. Boksjø (loc. 3). On the other hand *Psectrocladius (Monopsectrocladius) septentrionalis* Chern. made up about 2/3 of the orthoclad fauna in Lake N. Boksjø and only about 2 % in Lake S. Boksjø. *Psectrocladius (P) sordidellus* gp. had an increase in abundance at loc. 3, but totally the genus decreased in the lake. *Parakiefferiella* cf. *bathophila* (Kieff.) also showed increased abundance in Lake S. Boksjø compared with Lake N. Boksjø, while *Chironomus* spp. had a slight decrease. *Phaenopsectra* sp. which was rather frequent in the unlimed lake almost disappeared from the limed one. Other species with increased abundance at loc. 3 were *Dicrotendipes tritomus* gp., *Pseudochironomus prasinatus* Staeg. and *Polypedilum* cf. *breviantennum* Chern.

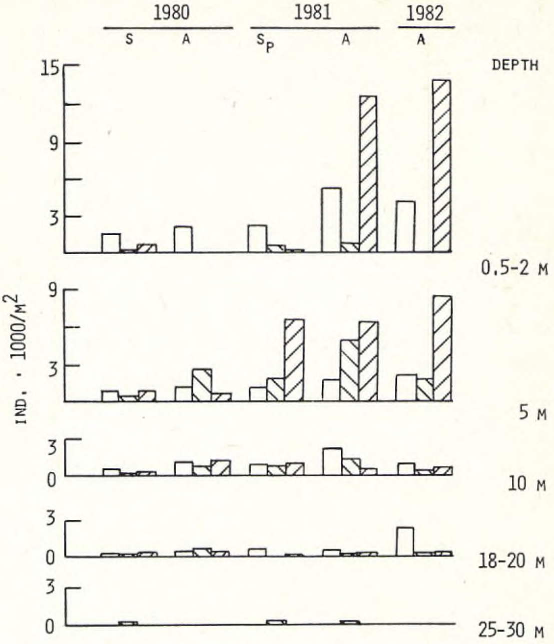


Fig. 4. Mean number of chironomids per m² at different depths.
□=station 1, ▧=station 2, ▨=station 3.
S=summer, A=autumn, Sp=spring.

A striking feature of Lake S. Boksjø is that the profundal of the lake is almost empty of chironomids from 10 m depth. Species present in the profundal in Lake N. Boksjø, but absent in this part in Lake S. Boksjø (loc. 3) are *Ablabesmyia monilis/phatta* (Egg.), *Procladius* sp., *Chironomus halophilus* gp., *D. tritomus* gp. and *P.* cf. *breviantennum*. *H. apicalis* was the only species well distri-

Table 4. *Number of chironomid species/species-groups per m² in Lakes N. Boksjø (1) and S. Boksjø (2) and (3).*

	Quantitative samples													Qualitative samples		
Station	1					2			3					1	2	3
Depths, meter	0.5	2	5	10	20	5	10	20–25	0.5	2	5	10	20–25	0–0.5		
TANYPODINAE																
Ablabesmyia monilis (L.)/*phatta* (Egg.)	175	35	140	—	120	40	—	—	120	475	240	—	—	—	—	—
Arctopelopia sp.	—	125	—	—	—	—	—	—	60	—	—	—	—	+	+	+
Macropelopia goetghebueri (Kieff.)	—	—	—	—	—	—	—	—	60	40	40	—	—	—	—	—
Procladius (*Holotanypus*) sp.	—	320	—	—	240	250	—	—	770	1640	445	—	—	—	—	—
Thienemannimyia sp.	35	70	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Tanypodinae indet.	35	—	—	30	40	—	—	—	—	300	—	—	—	—	—	—
ORTHOCLADIINAE																
Heterotanytarsus apicalis (Kieff.)	—	—	—	—	40	—	—	—	1240	1465	520	570	40	—	—	—
Heterotrissocladius grimshawi (Edw.)	—	—	—	30	—	—	—	—	60	—	—	50	—	—	+	+
Parakiefferiella cf. *bathophila* (Kieff.)	105	35	—	—	40	80	—	—	2770	170	40	—	40	—	—	+
Psectrocladius (*Monopsectrocladius*) *septentrionalis* Chern.	1505	295	290	—	—	—	—	30	120	40	80	—	—	—	—	—
Psectrocladius (*Psectrocladius*) *psilopterus* gp.	—	—	—	—	—	—	—	—	—	—	—	—	—	+	+	+
Psectrocladius (*Psectrocladius*) *sordidellus* gp.	385	—	95	—	210	250	—	30	2830	1120	610	—	40	—	—	+
Zalutschia cf. *tornetraeskensis* (Edw.)	—	—	—	—	—	—	—	—	60	—	—	—	—	—	—	—
Zalutschia zalutschicola Lipina	—	—	45	30	—	—	—	—	—	—	—	—	—	—	—	—
Orthocladiinae indet.	—	—	—	—	—	—	—	—	170	40	—	—	—	—	—	—
CHIRONOMINI																
Chironomus annularius gp.	—	35	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Chironomus halophilus gp.	—	100	—	—	40	120	—	—	—	1165	650	—	—	—	—	—
Chironomus thummi gp.	—	915	—	—	—	—	—	—	—	560	200	—	—	—	—	—
Cladopelma laccophila gp.	—	—	95	—	—	—	—	—	—	—	—	—	—	—	—	—
Cryptochironomus sp.	—	—	—	—	—	—	—	—	—	85	—	—	—	—	—	—
Demicryptochironomus vulneratus (Zett.)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—
Dicrotendipes tritomus gp.	70	70	45	—	420	80	—	—	120	2285	2875	—	—	+	—	—
Pagastiella orophila (Edw.)	175	680	990	30	630	280	—	—	300	1175	610	50	—	—	—	—
Phaenopsectra sp.	105	620	—	330	—	—	—	—	60	40	—	—	—	—	—	—
Polypedilum bicrenatum gp.	—	70	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Polypedilum cf. *breviantennum* Chern.	—	190	185	450	340	280	210	—	600	5515	1415	—	—	—	+	+
Polypedilum nubeculosum gp.	525	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—
Pseudochironomus prasinatus (Staeg.)	35	—	—	—	—	40	—	—	1530	950	200	—	—	+	—	+
Stictochironomus cf. *rosenschoeldi* (Zett.)	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+	—
Tribelos intextus (Walk.)	70	70	—	—	—	—	—	—	—	165	40	—	—	—	—	—
Chironomini indet.	105	—	—	—	—	—	—	—	—	40	—	—	—	—	—	—
TANYTARSINI																
Cladotanytarsus mancus gp.	—	—	—	—	—	—	—	—	60	—	—	—	—	—	—	+
Tanytarsus chinyensis gp.	—	—	—	—	—	—	—	—	—	—	—	—	—	—	—	+
Tanytarsus pallidicornis gp.	—	—	—	—	—	—	—	—	120	—	—	—	—	—	—	+
Tanytarsus sp.	35	35	45	—	—	—	—	—	350	40	40	—	—	—	—	—
SUM	3360	3605	1930	900	2120	1420	210	60	11400	17310	8015	670	120			

buted in the profundal in the autumn 1982 in Lake S. Boksjø.

To support the tendency of a decreasing number of species in the deeper part of the limed lake the spring data from 1981 will be given. At this time loc. 3 at 10 m depth had 924 chironomids per m². The taxa represented were *A. monilis/phatta, Heterotrissocladius grimshawi* (EDW.), *Mesocricotopus thienemanni* (GOETGH.), *Psectrocladius (M.) septentrionalis, Dicrotendipes tritomus* gp., *Stictochironomus* cf. *rosenschoeldi* (ZETT.) and *Tanytarsus* sp. *S.* cf. *rosenschoeldi* represented at that time about 44 % of the total chironomid fauna. In the autumn 1982 this species was present in qualitative shore samples only.

Many factors may be responsible for the observed "cut" below 5 m depth in the chironomid community. One important factor may be found in water-chemistry processes after liming. DICKSON (1983) and WRIGHT and SKOGHEIM (1983) have found that Al-complexes precipitate with increasing pH and accumulate in the sediment. This accumulation will probably be largest in the profundal zone. Such Al-compounds are shown to be toxic to fish (BAKER and SCHOFIELD 1980, MUNIZ and LEIVESTAD 1980, SKOGHEIM *et al.* 1984). ENGBLOM and LINGDELL (1983) suggested that they also affected the ephemerid fauna in running water during episodes of increasing pH.

These toxic effects might also be responsible for the negative development of the chironomid society in the profundal compared to the littoral/sublittoral zone.

Mayflies and caddisflies were also present in the quantitative samples in the littoral zone. The number of *Leptophlebia* sp. was highest in the autumn 1981 with about 2000 larvae per m² at loc. 3. All the other measurements showed values between 0 and 600 ind. per m².

V. REFERENCES

ALMER, B., W. DICKSON, C. EKSTRÖM and E. HÖRNSTRÖM. 1978. Sulfur pollution and the aquatic ecosystem. p. 271—311. *In* Sulfur in the environment. Part II. Ecological impacts. Ed.: J. O. Nriagu. John Wiley and Sons. New York.

BAKER, J. and C. L. SCHOFIELD. 1980. Aluminium toxicity to fish as related to acid precipitation and Airondack surface water quality. p. 292—293. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

BORGSTRØM, R., J. A. EIE, G. HARDENG, R. NORDBAKKE, J. E. RAASTAD and J. O. SOLEM. 1974. Investigations of potential national parks in Østfold County. Lab. Freshw. Ecol. Inland Fish., Oslo. Rep. 17: 1—71 (In Norwegian.)

BRITTAIN, J. E. and A. LILLEHAMMER. 1978. The fauna of the exposed zone of Øvre Heimdalsvatn: Methods, sampling stations and general results. Holarct. Ecol. 1: 221—228.

DICKSON, W. 1983. Liming toxicity of aluminium to fish. Vatten 39: 400—404.

DRABLØS, D. and A. TOLLAN. (Eds.) 1980. Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. SNSF project, Oslo-Ås. 383 p.

DRIVER, E. A. 1981. Calorific values of pond invertebrates eaten by ducks. Freshw. Biol. 11: 579—581.

ENGBLOM, E. and P.-E. Lingdell. 1983. The benthic fauna as pH-indicator. Rep. Nat. Swedish Environm. Protec. Bd SNV PM 1741. 181 p. (In Swedish with English abstract.)

ERIKSSON, F., E. HÖRNSTRÖM, P. MOSSBERG and P. NYBERG. 1983. Ecological effects of lime treatment of acidified lakes and rivers in Sweden. Hydrobiologia 101: 145—164.

FROST, S., A. HUNI and W. E. KERSHAW. 1971. Evaluation of a kicking technique for sampling stream bottom fauna. Can. J. Zool. 49: 167—173.

HENRIKSON, L., H. G. NYMAN, H. G. OSCARSON and J. A. E. STENSON. 1981. Rotatorieutvecklingen i en försurad sjö efter behandling med kalciumhydroxid, $Ca(OH)_2$. Inst. Zool. Univ., Gothenburg. 5 p. (In Swedish.)

HULTBERG, H., and I. ANDERSSON. 1981. Liming of acidified lakes and streams — aspects on induced physical-chemical and biological changes. Swed. Wat. Air Poll. Res. Inst., Gothenburg. IVL. Publ. B 621. 66 p.

KALKINGSPROSJEKTET 1983. Årsrapport 1980/81. Kalkingsprosjektet i Gjerstad, Aust-Agder. Rapp. 6/83: 1—101. (In Norwegian.)

MATTHIAS, U. 1982. Der Einfluss der Wasserstoffionenkonzentration auf die Zusammensetzung von Bergbachbiocoenosen, dargestellt an einigen Mittelgebirgsbäcken des Kaufunger Waldes (Nordhessen/Südniedersachsen). Universität des Landes Hessen, Kassel. 135 p.

MOSSBERG, P. and P. NYBERG. 1979. Bottom fauna of small acid forest lakes. Rep. Inst. Freshw. Res., Drottningholm 58: 77—87.

MUNIZ, I. P. and H. LEIVESTAD. 1980. Toxic effects of aluminium on the brown trout, *Salmo trutta* L. p. 320—321. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo-Ås.

RADDUM, G. G. 1979. Effects of low pH on insect larvae. SNSF Project IR 45/79. Oslo—Ås. 58 p. (In Norwegian with English summary.)

— and A. FJELLHEIM. 1984. Acidification and early warning organisms in freshwater in Western Norway. Verh. intern. Verein. Limnol. 22: 1973—1980.

— and O. A. SÆTHER. 1981. Chironomid communities in Norwegian lakes with different degrees of acidification. Verh. intern. Verein. Limnol. 21: 399—405.

SCHOFIELD, C. L. 1976. Acid precipitation: Effects on fish. Ambio 5: 228—230.

SKOGHEIM, O. K., B. O. ROSSELAND and I. SEVALDRUD. 1984. Deaths of spawners of Atlantic Salmon in River Ogna, SW Norway, caused by acidified aluminium-rich water. Rep. Inst. Freshw. Res., Drottningholm 61: 195—202.

STENSON, J. A. E. 1979. Predator — prey relations between fish and invertebrate prey in some forest lakes. Rep. Inst. Freshw. Res., Drottningholm 58: 166—183.

SUTCLIFFE, D. W. and T. R. CARRICK. 1973. Studies on mountain streams in the English Lake District. Freshw. Biol. 3: 437—462.

SWEDISH MINISTRY OF AGRICULTURE. 1982. Acidification today and tomorrow. The 1982 Stockholm conference on the acidification of the environment. 231 p.

THÖRNELÖF, E. 1982. Arbetsrapport från födovalsanalys och åldersbestämning av 1980 års material på fisk från kalkningsprojekt Boksjö-Kornsjö, Enningdalsälvens vattensystem. (Working report of food and age analysis of fish, caught in 1980 in the Lakes Boksjö and Kornsjö, the watershed of Enningdalsälven. Fiskeriintendenten i västra distriktet, Göteborg. 25 p. (Mimeographed in Swedish.)

WIEDERHOLM, T. and L. ERIKSSON. 1977. Benthos of an acid lake. Oikos 29: 261—267.

WRIGHT, R. F. and O. K. SKOGHEIM. 1983. Aluminium speciation at the interface of an acid stream and a limed lake. Vatten 39: 301—304.

A Comparative Study on Salmonid Fish Species in Acid Aluminium-Rich Water

I. Mortality of eggs and alevins

ODD K. SKOGHEIM and BJØRN OLAV ROSSELAND

Directorate for Wildlife and Freshwater Fish, Fish Research Division, P.O. Box 63, N-1432 Ås-NLH, Norway

ABSTRACT

Eggs of Atlantic salmon, sea trout, brown trout and brook trout were fertilized and reared in two different waters: lake water pH 6.4—6.8, calcium 3—4.5 mg/l, and labile Al < 10 µg/l, and acid brook water pH 4.8—5.6, calcium 1—2.5 mg/l and labile Al 20—310 µg/l. Due to species differences in spawning time (October 26 to November 29), as well as seasonal changes in water quality, pH and Al levels to which developmental stages were exposed varied among species. Only sea trout and brown trout eggs were spawned simultaneously and were exposed to comparable water quality throughout their development. For all species, few fish died in lake water (1.1—5.4 %) through the end of the alevin stage. In acid brook water, the corresponding mortality was 99.7 % for sea trout, 93.2 % for salmon, 86.9 % for brown trout and 12.9 % for brook trout. For sea trout 97 % of the mortality occurred during hatching, compared to 57, 35 and 8 % mortality during hatching for brown trout, salmon and brook trout respectively. Differences in hatching mortality seemed to be species dependent, but also correlated to differences in pH and aluminium. Due to differences in the timing of hatch, mean pH varied from 4.93 to 5.15 and mean concentrations of labile Al from 93 to 183 µgAl/l during hatching periodes for the different species. Highest Al concentrations corresponded to the lowest pH. Degree days, from fertilization to initiation of feeding, were significantly greater in the acid brook water than in the lake water, principally as a result of retardation of development of the alevin stage. The increase in time from hatch to initiation of feeding in brook water relative to lake water was 80, 41, 41 and 19 % for salmon, sea trout, brown trout and brook trout, respectively.

I. INTRODUCTION

Acidification of rivers and lakes is the most serious threat to freshwater fisheries in Norway. Lakes and rivers in an area of 33,000 km² are currently affected (SEVALDRUD *et al.* 1980). Historically, acid water has been recognized as a problem in the southern counties (Sørlandet) since the 1920s (DAHL 1927), with the first observations of fish kills attributed to acidity in 1911 (HUITFELDT-KAAS 1922). Acid water soon caused decreased production of eggs and fry of Atlantic salmon (*Salmo salar* L.) and brown trout (*Salmo trutta* L.) in hatcheries (SUNDE 1926, DAHL 1927), resulting in the installation of limestone and shellsand filters as common practice (BAKKE 1939). Today, salmon have been eliminated from all major rivers in southernmost Norway (Sørlandet) (JENSEN and SNEKVIK 1972, LEIVESTAD *et al.* 1976).

Atlantic salmon was the first species affected in Norwegian waters. Likewise, in laboratory experiments salmon have been found to be especially sensitive to acid water (BUA and SNEKVIK 1972, JOHANSSON *et al.* 1977, GRANDE *et al.* 1978). The youngest stages (eggs — fingerlings), in particular, have been identified as most sensitive (JENSEN and SNEKVIK 1972, LEIVESTAD *et al.* 1976). Several studies have been conducted on the effect of pH (H^+) on oogenesis, embryonic development, hatching and absorption of the yolk sac (in the alevin stage) (see the extensive review by PETERSON *et al.* 1982). In recent years investigators have recognized that acid waters are associated with elevated levels of aluminium (Al), and these Al concentrations have been shown to have a deleterious effect on both 0+ and older fish (SCHOFIELD 1977, BAKER and SCHOFIELD 1980, MUNIZ and LEIVESTAD 1980).

The present study was performed using eggs of Atlantic salmon, sea trout, brown trout and brook trout exposed to two types of water, lake water with pH 6.4—6.8 and brook water with pH 4.8—

5.6. Another experiment, using older stages of the same species, was performed in parallel, and is described in ROSSELAND and SKOGHEIM (1984).

II. MATERIAL AND METHODS

All experiments were conducted during the period October 1981 to June 1982 at the Research Station for Freshwaterfish situated at the outlet of River Imsa, SW Norway.

Two water sources were used; water from River Imsa taken at the outlet of Lake Livatn (lake water, LW), circumneutral and well buffered, and water from the acid brook Fossbekken (brook water, BW) situated within the River Imsa catchment area. Before entering the hatchery, both waters were aerated and the temperature of the brook water was adjusted to match that of the lake water.

Four groups were tested, representing eggs of three salmonid species: 1 Atlantic salmon (*Salmo salar* L.) (Imsa strain), 2 sea trout (*Salmo trutta* L.) (Imsa strain), 2 brown trout (*Salmo trutta* L.) (Fossbekken strain) and 3 brook trout (*Salvelinus fontinalis* MITCHILL). Eggs were collected from seatrout and brown trout on October 26, from brook trout on November 10, and from salmon on November 29. All eggs were fertilized and swelled in lake water. After fertilization, the eggs were placed in two flow-through hatching channels (Ewos, mod. 2003) each holding 4 separate boxes. The residence time of water in the channels was about 40 minutes. Both channels were supplied with lake water until November 29 at which time, following fertilization of salmon eggs, one of the channels was switched over to the acid brook water. The channels were checked daily for mortality, and dead eggs and fry were removed. Dead eggs were checked for fertilization success and development. Only fertilized eggs were used in the calculation of cumulative mortality. Survival was monitored through the end of the alevin stage (initiation of feeding): through April 1982 for sea trout, brown trout and brook trout; through June 1982 for salmon. Temperature was measured daily in both channels. Water samples were taken daily from the channel with brook water and weekly from the channel with lake water. Aliquots of water samples were immediately prepared for aluminium analyses. Samples were passed through a cation-exchange column (DOWEX-50, 20/50 mesh, prepared with Na^+) and analysed by the pyrocatechol violet method (DOUGAN and WILSON 1974). This fraction is called non-labile Al and is assumed to include primarily organically complexed monomeric aluminium. The aluminium fraction analysed directly without any pretreatment is termed acid-reactive Al and the difference between these two fractions is labile Al and is assumed to include inorganic monomeric species of aluminium. The latter fraction is considered to include the toxic Al forms (DRISCOLL *et al.* 1980). This fractionation scheme is a slight modification of that presented by DRISCOLL (1980).

pH was measured potentiometrically with a Radiometer PHM 64. Calcium, magnesium, sodium, potassium, iron and manganese were determined by atomic absorption spectrophotometry (Perkin Elmer Mod. 603). Zinc, copper, lead and cadmium were determined by potentiometric stripping analyses with Radiometer ISS 820. Alkalinity was determined by automatic titration with a Radiometer RTS 822. Sulphate and chloride were analysed by an ion exchange technique combined with conductometry (modified after STAINTON 1974). Total fluoride was analysed using an ion selective electrode (ORION 1976). Conductivity was measured with a Hoelze & Chelius conductivity meter model L17 and given at 25°C.

Dissolved organic matter was estimated by UV absorbance (UVICORD LKB 8300) at 254 nm with a 1 cm cuvette. Sporadically, dissolved organic matter was estimated against a Pt standard by means of a comparator, and as chemical oxygen demand ($KMnO_4$ oxidant).

III. RESULTS

Temperature and water quality

In Fig. 1 A, temperature variations in the two hatching channels are shown. Temperatures in the brook water were adjusted to match with natural fluctuations in the lake water. During December, temperatures in the two channels differed at most 2°C. From mid January through April, the tem-

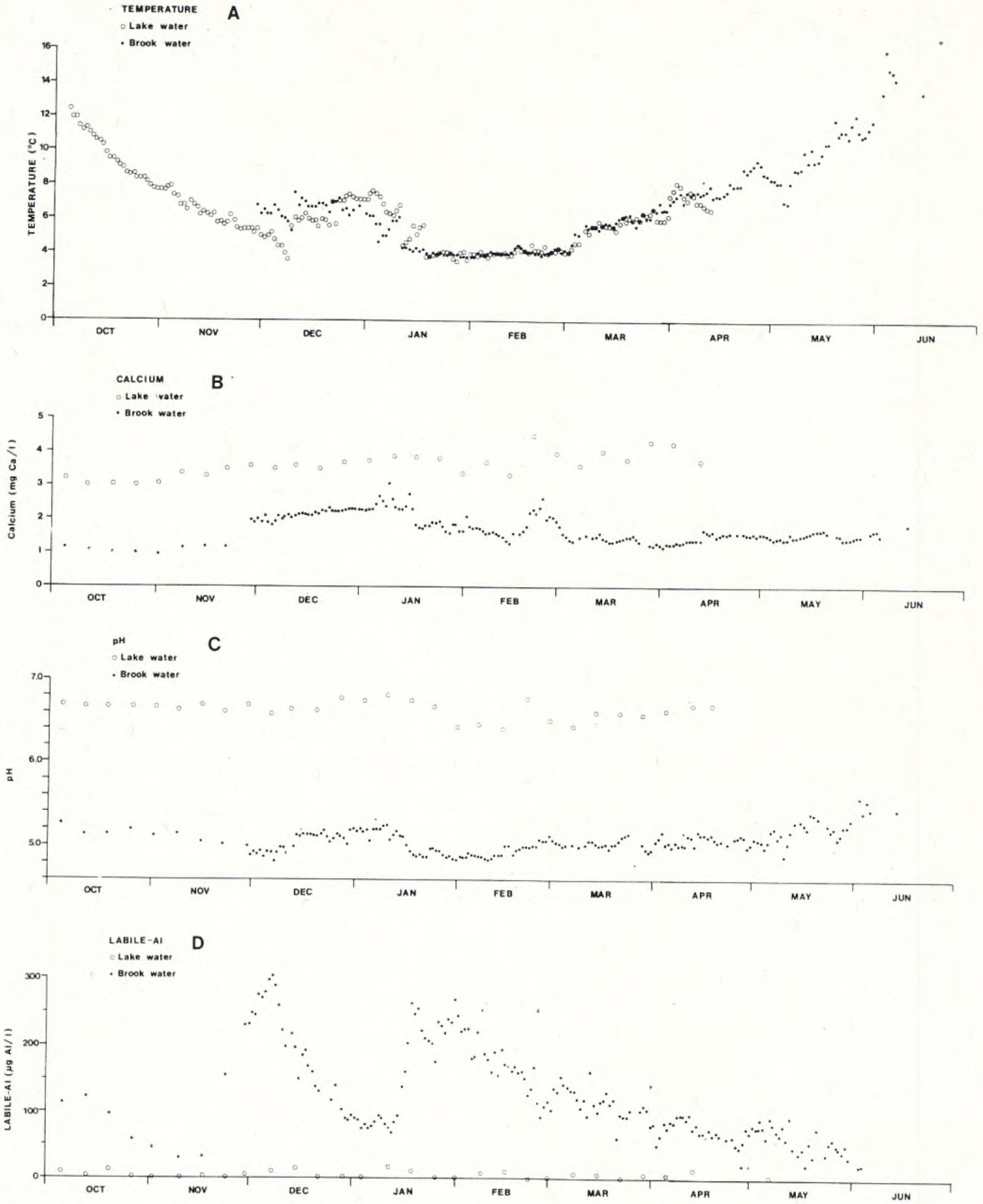


Fig. 1. Variations in physical and chemical characteristics for lake water (LW) and brook water (BW) during the experimental period. A. Temperature. B. Calcium. C. pH. D. Labile-Al.

perature differences were even smaller. Temperatures in the two channels increased steadily from 4°C in mid January to 16°C by mid June. Variations in pH and calcium in lake water and brook water are shown in Figs. 1 B and 1 C and Table 1. The lake water was circumneutral with pH varying from 6.4 to 6.8. The concentration of calcium varied between 3.0 and 4.5 mg Ca/l and the average alkalinity was 0.09 meq/l. Chemical concentrations varied little in the lake water during the experimental period. The brook water was acid with pH between 4.8 and 5.6. The con-

Table 1. *Water quality of brook water and lake water during the experimental period. Maximum (Max) and minimum (Min) and number of observations (n) are given.*

		Brook water		Lake water	
		Max—Min	n	Max—Min	n
pH		5.59— 4.82	178	6.77— 6.44	25
Conductivity	µs/cm	88.5 —36.0	193	84.0 —61.0	25
Calcium	mgCa/l	3.10— 0.94	193	4.50— 3.07	25
Magnesium	mgMg/l	1.42— 0.95	79	1.38— 1.26	11
Sodium	mgNa/l	11.4 — 7.4	79	7.5 — 6.6	11
Potassium	mgK/l	6.3 — 0.39	79	3.65— 1.70	11
Sulphate	mgSO$_4$/l	4.9 — 2.9	13	6.5 — 5.1	8
Chloride	mgCl/l	13.7 —10.3	13	13.6 —11.7	8
Alkalinity	meq/l	< 0.010	15	0.100—0.067	9
Iron	mgFe/l	0.06—< 0.01	79	0.03—< 0.01	11
Manganese	mgMn/l	0.07— 0.02	79	0.02—< 0.01	11
Acid reactive Al	µgAl/l	375—72	180	65—43	20
Non-labile Al	µgAl/l	84—36	176	60—30	20
UV-absorbance	cm^{-1}	0.128—0.042	176	0.088—0.059	21

centration of calcium varied between 1.1 and 3.1 mg Ca/l. The alkalinity was 0 meq/l. Both pH and calcium varied considerably, associated principally with variations in weather conditions, such as intensity and composition of precipitation and duration of periods of melting/freezing in the catchment area. Concentrations of other major chemical constituents were relatively stable during the experimental period, Table 1.

Conductivity in lake water varied between 69 and 84 µS/cm and in brook water between 50 and 88 µS/cm. The contribution of the H$^+$ ion to conductivity was about 5 µS/cm at most.

The concentration of manganese was always lower than 0.01 mg/l in the lake water and lower than 0.07 mg/l, with a mean value of 0.04 mg/l, in brook water. Correspondingly, concentrations of iron always were lower than 0.06 mg/l, with a mean value of 0.03 mg/l in brook water. For four random samples, concentrations in brook water of zinc varied between 15 and 24 µg/l, copper between 7 and 8 µg/l, lead levels were < 2 µg/l, and cadmium < 0.4 µg/l.

In Fig. 1 D, levels of labile Al are shown. The concentration of labile Al was always < 10 µg/l in lake water, as expected at that pH level. In brook water, however, the concentration of labile Al varied between 20 and 310 µg/l. Two marked peaks in labile Al occurred, one in the first week of December and a second in mid January. The concentration of non-labile Al was relatively constant in both waters, ranging from 36 to 65 µg/l in lake water and 34 to 111 µg/l, with a mean value of 65 µg/l in brook water for the experimental period. Concentrations of organic matter were low in both waters. The chemical oxygen demand was typically in the order 1.0 to 2.0 mg O/l, and level of humic material was typically about 5 mg Pt/l. UV-absorbance was measured on all samples during the experimental period and varied between 0.040 and 0.080 cm^{-1}.

Mortality and development

For all species, little mortality occurred in the acid brook water prior to hatching (Fig. 2). During hatching, sea trout experienced near 100 % mortality, with cumulative mortality increasing from 2.4 % at the start of hatch to 99.1 % at the end of the 3-week hatching period. Increase in cumulative mortality during the hatching period were less for the other species; brown trout 4.2 to 61.0 %; salmon 4.0 to 38.9 %; and brook trout 4.7 to 12.4 % respectively (Fig. 2). After hatching, mortality of alevins of salmon and brown trout continued, with cumulative mortality increasing from 38.9 to 93.2 % at the end of the experiment for salmon, and from 61.0 to 85 % for brown trout. Mortality of brook trout alevins (post-hatching), was negligible, with cumulative mortality increasing only from 12.4 to 12.9 %.

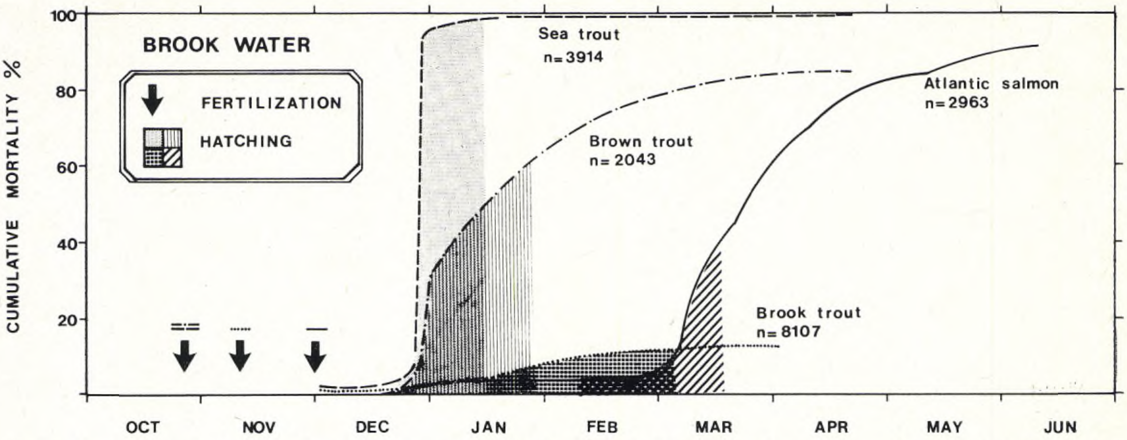


Fig. 2. Cumulative mortality as a function of time in the acid brook water. Fertilization time and hatching period are given, as are the number of eggs used in each group.

In lake water, few fish died during the experimental period (from fertilization to end of the alevin stage); for salmon total mortality was 1.1 %, sea trout 5.1 %, brown trout 3.1 % and brook trout 5.4 %. Time of death was associated with neither hatching nor resorption of the yolk sac.

Exposure to acid brook water was initiated at the same date (November 29) for all species. Due to the species difference in spawning time, however, the period of exposure corresponded to different stages of embryo development. Only sea trout and brown trout were fertilized simultaneously (Fig. 2) and thus experienced the same water quality and temperature throughout their development. As can be seen from Fig. 2, hatching for the two forms of trout began on the same date, but ended earlier for sea trout. Direct comparisons of effects of lake water and brook water on embryo survival, therefore are only valid for sea trout and brown trout during this initial period of the study. At the end of this hatching period, cumulative mortality for brown trout reached 45.3 % compared to 99.1 % for sea trout.

In Table 2, water quality and cumulative mortality during the hatching period are listed for each species.

Temperature is a major determinant of the rate of embryo development. Thus Fig. 3 indicates number of degree days to three developmental stages (eyed eggs, hatch and initiation of feeding) for each species in the two waters. Time to hatching was not significantly different in the two waters, but for each species initiation of feeding was delayed in the acid brook water relative to the lake water.

Table 2. *Water quality and cumulative mortality during hatching for three salmonid fish species including two forms of trout. Mean (M), standard deviation (SD), maximum (Max) and minimum (Min) values of pH, calcium and labile Al are given.*

	pH		Calcium, mg Ca/l		Labile Al, µg Al/l		Cummulative mortality during hatching, %
	M ± SD	Max—Min	M ± SD	Max—Min	M ± SD	Max—Min	Start → end
Atlantic salmon	4.99 ± 0.08	5.13—4.85	1.71 ± 0.28	2.35—1.31	142 ± 29	191—93	4.0 → 38.9
Sea trout	5.15 ± 0.06	5.25—5.05	2.35 ± 0.19	3.10—2.23	93 ± 19	139—74	2.4 → 99.1
Brown trout	5.08 ± 0.12	5.25—4.82	2.24 ± 0.27	3.10—1.74	130 ± 64	263—69	4.2 → 61.0
Brook trout	4.93 ± 0.09	5.18—4.85	1.85 ± 0.29	2.76—1.37	183 ± 47	269—84	4.7 → 12.4

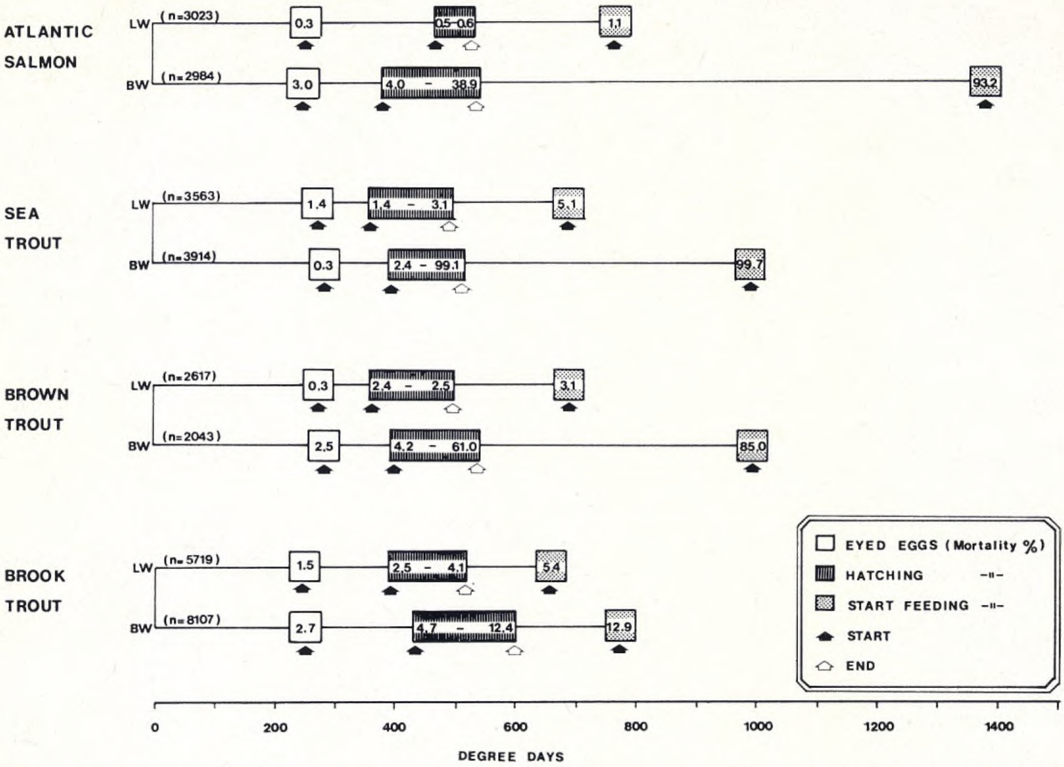


Fig. 3. Embryonic stage as a function of degree days (days × temperature (°C)) in the two waters. Start and end (hatching only) for the different stages are given, with the corresponding cumulative mortality (%). The number (n) of eggs in each group is given in parantheses.

IV. DISCUSSION

The concentration of labile Al varied considerably during the experimental period (Fig. 1). Variations in pH between 4.8 and 5.6 (and associated changes in the concentration of the OH^- ligand) may to a large extent account for this variation. Variations of other organic ligands, *e.g.*, flouride, may also be of importance. Seip *et al.* (1984) stressed the temperature dependency of solubility constants for gibbsite ($Al(OH)_3$). Solubility of gibbsite may change by a factor of 10 within the temperature range measured during the experiment. Temperature variations may also have strongly influenced the concentration of labile Al. In order to evaluate the importance of pH and temperature in controlling observed concentrations of labile Al, both simple and multiple correlation analyses using labile Al and log (labile Al) as the dependent variable were performed. The multiple regression log (labile Al) = f (pH, temperature) provided the best model. The variation in pH and temperature explained 66 % of the variation in log (labile Al) (based on 158 observations).

Brook water used in these experiments was aerated and passed through foam-plastic prior to entering the hatching channels. Measured inorganic aluminium concentrations are thus not likely to have exceeded equilibrium with the solid phase.

Aluminium speciation was calculated for samples collected over a 3-day period in February 1982 (Rosseland and Skogheim 1984). Based on the solubility constant of a solid $Al(OH)_3$ phase corresponding to that of synthetic gibbsite (May *et al.* 1979), aluminium levels in brook water were slightly undersaturated and the dominating forms were aqueous Al^{3+}, $Al(OH)^{2+}$, $Al(OH)_2^+$ and AlF^{2+}. Calculations of the aluminium speciation for the whole experimental period will be reported elsewhere.

Mortalities occurred principally during and after hatching for all species. The cumulative mortality through initiation of feeding was 99.7 % for sea trout, 93.2 % for salmon and 85 % for brown trout, but only 12.9 % for brook trout. Mortality during hatching was highest for sea trout and brown trout, whereas mortality during the alevin stage was highest for salmon. As salmon are generally considered more acid-sensitive than sea trout or brown trout, the lower hatching mortality in spite of a lower pH during hatching (Table 2) is surprising. Interspecies comparisons, however, are difficult because of species differences in spawning time and rate of development in conjunction with variations in water quality through time.

High mortality during hatching have been observed by several authors (RUNN *et al.* 1977, PETERSON *et al.* 1980). The special susceptibility at this stage has been attributed to inactivation of the "hatching enzyme" (chorionase) caused by a reduction in pH in the perivitelline fluid (PETERSON *et al.* 1980, JOHANSSON *et al.* 1981, HAYA and WAIWOOD 1981). Species and form difference in effects of acid brook water on hatching may thus reflect variations among species in levels of inactivation of the hatching enzyme.

Aluminium toxicity may also be important. BAKER and SCHOFIELD (1980) showed that for eggs of brook trout, aluminium concentrations up to 500 µg Al/l improved survival at pH less than 4.6 to 4.9 (dependent on Al level), but decreased survival through hatching at pH 5.0 to 5.5. For alevins, aluminium had the least negative effect at pH 4.6—4.9. Mortality attributable to aluminium increased with increasing pH to 5.5, especially in oversaturated Al solutions. Using the same levels of aluminium and a calcium level of 2 mg Ca/l, BROWN (1983) demonstrated a similar response for alevins of brown trout in the pH range 4.5 to 5.4, the highest mortality being at pH 5.4.

In our experiments alevin mortality for brown trout occurred in the period late January until mid March; pH was relatively stable but steadily increasing, varying between 4.9 to 5.0 with concentrations of labile Al decreasing from 250 to 100 µg Al/l. Alevin mortality for salmon occurred mid March to end of May, while pH increased from about 5.0 to 5.5 and labile Al decreased from about 125 to 25 µg Al/l. Results for eggs and alevins for these salmonid species generally fit those of BAKER and SCHOFIELD (1980) and BROWN (1983), although data for comparison are limited for any one species and strain.

Other complicating factors in our experiments are the levels of copper and zink. Although the concentrations are lower than the toxic levels described by ALABASTER and LLOYD (1982) and BAKER (1982), no continuous sampling for heavy metals in connection to changes in pH in the brook water were performed. We can therefore not exclude joint effects of these metals in combination with pH and aluminium.

Temperature has also been reported as an important determinant of egg (embryo) and fry mortality. For rainbow trout, *Salmo gairdneri*, in acid water, resistance to low pH decreased at low temperature (KWAIN 1975). Differences in temperatures during exposure periods may therefore also have influenced the apparent relative species and form sensitivity to acid brook water.

Temperature is a main factor in determining rate of embryo development, expressed as degree days until hatching and resorption of yolk sac (Fig. 3). Exposure to acid brook water delayed development in all species with the effect occurring principally at the alevin stage. The increase in degree days was greatest for salmon, requiring 80 % more degree days to initiation of feeding in the acid water. Differences in degree days were 45, 45 and 18 % for sea trout, brown trout and brook trout, respectively. A similar delay in development, especially during hatching, has been reported by others (RUNN *et al.* 1977, PETERSON *et al.* 1980). Salmon eggs began hatching earlier in acid brook water than in lake water, a phenomenon also reported by TROJNAR (1977) and CARRICK (1979).

Prolonged development time for alevins in acid brook water may reflect a decrease in metabolism. This is in contrast to the observed increase in metabolism for adult brook and brown trout in acid aluminium-rich water reported by ROSSELAND (1980). A decrease in metabolism in acid water, and thus a prolonged development time through initiation of feeding and swim-up, may decrease the probability that post swim-up fry are exposed to acid episodes during snowmelt.

V. ACKNOWLEDGMENTS

We gratefully acknowledge the assistance given by the staff at the Research Station for Freshwaterfish, Ims, especially TORMOD HUSABØ and CHRISTOFFER SENSTAD, and the staff at the laboratory at Ås, especially EDEL OPEM.

A special thank to the Drs. JOAN BAKER, MAGNE GRANDE, ARNE HENRIKSEN, KJELL W. JENSEN and HELGE LEIVESTAD for valuable comments and improvements of the manuscript.

VI. REFERENCES

ALABASTER, J. S. and R. LLOYD. 1982. Water quality criteria for freshwater fish. FAO/Butterworths, London — Boston. 361 p.

BAKER, J. P. 1982. Effects on fish of metals associated with acidification. p. 165—176. *In* Acid rain/fisheries. Ed.: R. E. Johnson. Proc. Int. Symp. Acidic Rain and Fishery Impacts on Northeastern North America. Am. Fish. Soc., Bethesda, Maryland.

— and C. L. SCHOFIELD. 1980. Aluminium toxicity to fish as related to acid precipitation in Adirondack surface water quality. p. 292—293. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF project, Oslo—Ås.

BAKKE, J. 1939. Klimaforandring og laksebestand. (Climatic changes and salmon stocks.) Fiskesport 10: 201—203. (In Norwegian.)

BROWN, D. J. A. 1983. Effects of calcium and aluminium concentration in the survival of brown trout (*Salmo trutta*) at low pH. Bull. Environm. Contam. Toxicol. 30: 582—587.

BUA, B. and E. SNEKVIK. 1972. Klekkeforsøk med rogn av laksefisk 1966—1971. Virkning av surhet og saltinnhold i klekkevannet. (Hatching experiments with roe from salmonid fishes 1966—1971. Effects of acidity and dissolved salt in the water.) Vann 7: 86—93. (In Norwegian.)

CARRICK, T. R. 1979. The effect of acid water on the hatching of salmonid eggs. J. Fish. Biol. 14: 165—172.

DAHL, K. 1927. The effect of acid water on trout fry. Salm. Trout Mag. 46: 35—43.

DOUGAN, W. K. and A. L. WILSON. 1974. The absorptiometric determination of aluminium in water. A comparison of some chromogenic reagents and the development of an improved method. Analyst, Camb. 99: 413—430.

DRISCOLL, C. T. 1980. Chemical characterization of some dilute acidified lakes and streams in the Adirondack Region of New York State. Ph. D. Thesis. Cornell Univ. 308 p.

— J. P. BAKER, J. J. BISOGNI JR. and C. L. SCHOFIELD. 1980. Effect of aluminium speciation on fish in dilute acidified waters. Nature, Lond. 284: 161—164.

GRANDE, M., I. P. MUNIZ and S. ANDERSEN. 1978. Relative tolerance of some salmonids to acid water. Verh. Int. Verein. Limnol. 20: 2076—2084.

HAYA, K. and B. A. WAIWOOD. 1981. Acid pH and chorionase activity of Atlantic salmon (*Salmo salar*) eggs. Bull. Environm. Contam, Toxicol. 27: 7—12.

HUITFIELDT-KAAS, H. 1922. Om aarsaken til massedød av laks og ørret i Frafjordelven, Helleelven og Dirdalselven i Ryfylke høsten 1920. (On the cause of mass death of salmon and trout in Frafjordelven, Helleelven and Dirdalselven in Ryfylke autumn 1920). Norsk Jæger- Fiskefor. Tidsskr. (1/2): 37—44. (In Norwegian.)

JENSEN, K. W. and E. SNEKVIK. 1972. Low pH levels wipe out salmon and trout populations in southern Norway. Ambio 1: 223—225.

JOHANSSON, N., P. RUNN and G. MILBRINK. 1977. Early development of three salmonid species in acidified water. Zoon, Uppsala 5: 127—132.

— P. RUNN and M. SOHTELL. 1981. Perivitelline pH of salmonide eggs in relation to ambient pH. Wat. Res. Bull. 17: 6, 994—999.

KWAIN, W. 1975. Effects of temperature on development and survival of rainbow trout, *Salmo gairdneri*, in acid water. J. Fish. Res. Board Can. 32: 493—497.

LEIVESTAD, H., G. HENDREY, I. P. MUNIZ and E. SNEKVIK. 1976. Effects of acid precipitation on freshwater organisms. p. 87—111. *In* Impact of acid precipitation on forest and freshwater ecosystems in Norway. Ed.: F. H. Brække. SNSF-project FR 6/76. Oslo—Ås.

MAY, H. M., P. A. HELMKE and M. L. JACKSON. 1979. Gibbsite solubility and thermodynamic properties of hydroxy-aluminium ions in aqueous solution at 25°C. Geochim. Cosmochim. Acta 43: 861—868.

MUNIZ, I. P. and H. LEIVESTAD. 1980. Toxic effects of aluminium on the brown trout, *Salmo trutta* L. p. 320—321. *In* Ecological impact of acid precipitation. Proc. Inst. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

ORION, 1976. Instruction manual. Fluoride Electrodes. Orion Research Inc., Cambridge.

PETERSON, R. H., P. B. DAYE and J. L. METCALFE. 1980. Inhibition of Atlantic salmon, *Salmo salar*, hatching at low pH. Can. J. Fish. Aquat. Sci. 37: 770—774.

— R. G. DAYE, G. L. LACROIX and E. T. GARSIDE. 1982. Reproduction in fish experiencing acid and metal stress. p. 177—195. *In:* Acid rain/fisheries. Ed.: R. E. Johnson. Proc. Int. Symp. Acidic. Rain and Fishery Impacts on Northeastern North America. Am. Fish. Soc., Bethesda, Maryland.

ROSSELAND, B. O. 1980. Effects of acid water on metabolism and gill ventilation in brown trout, *Salmo trutta* L., and brook trout, *Salvelinus fontinalis* MITCHILL. p. 348—349 *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway

1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

— and O. K. Skogheim. 1984. A comparative study on salmonid fish species in acid aluminium-rich water. II. Physiological stress and mortality of one and two year old fish. Rep. Inst. Freshw. Res. Drottningholm 61: 186—194.

Runn, P., N. Johansson and G. Milbrink. 1977. Some effects of low pH on the hatchability of eggs of perch, *Perca fluviatilis* L. Zoon, Uppsala 5: 115—125.

Schofield, C. L. 1977. Acid snow-melt effects on water quality and fish survival in the Adirondack Mountains of New York State. Res. Proj. Tech. Comp. Rep. Project A-072-NY. Cornell University, Ithaca, New York. 27 p.

Seip, H. M., L. Müller and A. Naas. 1984. Aluminium speciation: Comparison of two spectrophotometric analytical methods and observed concentrations in some acidic aquatic systems in southern Norway. Wat. Air Soil Poll. 23: 81—95.

Sevaldrud, I. H., I. P. Muniz and S. Kalvenes. 1980. Loss of fish populations in Southern Norway. Dynamics and magnitude of the problem. p. 350—351. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

Stainton, M. P. 1974. An automated method for determination of chloride and sulphate in freshwater using cation exchanges and measurement of electric conductance. Limnol. Oceanogr. 19: 707—711.

Sunde, S. E. 1926. Surt vand dræper laks og ørretyngel. Norsk Jæger Fiskefor. Tidsskr. (2): 1—4.

Trojnar, J. R. 1977. Egg hatchability and tolerance of brook trout (*Salvelinus fontinalis*) fry at low pH. J. Fish. Res. Board Can. 34: 574—579.

A Comparative Study on Salmonid Fish Species in Acid Aluminium-Rich Water

II. Physiological stress and mortality of one- and two-year-old fish

BJØRN OLAV ROSSELAND and ODD K. SKOGHEIM

Directorate for Wildlife and Freshwater Fish, Fish Research Division, P.O. Box 63, N-1432 Ås-NLH, Norway

ABSTRACT

A comparative study between species, using one- and two-year-old Atlantic salmon, sea trout and brown trout and one-year-old brook trout, was performed in a flowthrough system at pH 4.9—5.0 with four different concentrations of labile aluminium (130—462 μg/l). Mortality (in 64 hrs) was dependent on species, age and aluminium concentration. Mortality increased with increased concentrations of aluminium-hydroxides. In all treatments, two-year-old (presmolt) salmon were most sensitive, and brook trout most resistant. Among the two-year-old fish, loss of plasma chloride was most rapid in the salmon, followed by sea trout and brown trout. Brook trout had an initial ion-loss comparable to the salmon, but the plasma ion concentration stabilized or increased after 10—20 hrs at all concentrations of aluminium. Except for brook trout, the hematocrit increased with a decrease in plasma ions. The result, illustrating the high sensitivity of salmon smolts, explains why episodic fish kills in the acidified River Vikedal (1981—83) affect primarily salmon smolts and not the younger year classes.

I. INTRODUCTION

Atlantic salmon (*Salmo salar* L.) are now extinct in all major rivers in southernmost Norway (Sørlandet) (JENSEN and SNEKVIK 1972, LEIVESTAD *et al.* 1976, MUNIZ 1981) and the problem seems to be spreading northward along the western coast of Norway. Experiments have demonstrated that Atlantic salmon are particularly sensitive to acid waters (BUA and SNEKVIK 1972, JOHANSSON *et al.* 1977, GRANDE *et al.* 1978), thus accounting for the fact that salmon is the first species to disappear from acidified rivers.

For salmon and other species, recruitment failure, resulting from high mortality of the youngest stages (egg-fingerling) has been considered as the main cause for population decline (JENSEN and SNEKVIK 1972, LEIVESTAD *et al.* 1976, ROSSELAND *et al.* 1980). Only in special cases have fish kills involving older fish been observed (HUITFELDT-KAAS 1922, ROSSELAND 1953, LEIVESTAD and MUNIZ 1976, SKOGHEIM *et al.* 1984, HESTHAGEN and SKOGHEIM 1984). After hatching, mortality seems to be dependent mainly on the concentrations of H^+ (pH), aluminium (Al) and calcium (Ca) (SCHOFIELD 1977, BAKER and SCHOFIELD 1980, MUNIZ and LEIVESTAD 1980, BROWN 1983). Apparently only inorganic species of aluminium are toxic (DRISCOLL *et al.* 1980). Recently, several reviews describing the toxicity of acid waters have been published, *e.g.*, BAKER (1982), WOOD and MCDONALD (1982).

During the 1981 snow melt in River Vikedal, western Norway, a fish kill involving mainly smolts of salmon, and to a less extent sea trout, (*Salmo trutta* L.), was observed. Younger year classes appeared unaffected. This observation was confirmed during similar fish kills in the spring of 1982 and 1983 (HESTHAGEN and SKOGHEIM 1984). Thus the higher vulnerability of the youngest stages was questioned. Experiments were conducted to determine the sensitivity of salmon smolt relative to younger salmon and other salmonid species. Data comparing one- and two-year-old Atlantic salmon, sea trout, brown trout, and one-year-old brook trout are presented here. The effects of acid waters on different age groups of Atlantic salmon, including eyed eggs, have been published separately (ROSSELAND and SKOGHEIM 1982). Parallel studies on early life stages of salmon, sea trout, brown trout and brook trout are discussed in SKOGHEIM and ROSSELAND (1984).

II. MATERIAL AND METHODS

The experiment was performed in February 1982 at the Research Station for Freshwaterfish situated at the outlet of River Imsa, SW Norway. Waters from two sources were used as in the experiment involving effects on development and hatching of eggs (SKOGHEIM and ROSSELAND 1984) in lake water and acid brook water. Three additional treatments were tested involving additions of aluminium (Al) to acid brook water. The experimental setup consisted of 4 flow-through tanks each with a volume of 100 l. The flow-through rate was 1 l/min.; water residence time of 100 min. per tank. To three of the tanks (B—D), aluminium (as a solution of $Al_2 (SO_4)_3$) was added; 120, 225 and 415 mg Al/l, respectively, via a peristaltic pump (Ismatec IP-12). Water and aluminium additions were mixed (by air-flushing) for 10 minutes prior to discharge into fish tanks. The experimental temperature was 5.2°C.

Fish used as controls were held in lake water in 800 l tanks (for one-year-old fish) and 6000 l tanks (for two-year-old fish). Experimental fish were transferred directly to acid water from lake water without any acclimation. Handling stress, however, was equal for all groups.

Three salmonid species including two forms of trout were used: Atlantic salmon (*Salmo salar* L.) (Imsa strain), sea trout (*Salmo trutta* L.) (Imsa strain), brown trout (*Salmo trutta* L.) (Randselv strain) and brook trout (*Salvelinus fontinalis* MITCHILL). Except for brook trout, for which only one-year-old fish were used, the other species were represented by both one- and two-year-old fish. The two-year-old Atlantic salmon and sea trout were undergoing smoltification (presmolt).

Mortality of each year class were recorded through the experiment (64 hrs). At intervals blood samples were drawn by heart puncture from the older fish. Samples were centrifuged immediately (MSE Micro hematrocrit centrifuge type 346, 3 min.) and analysed for hematocrit and plasma chloride (Radiometer CMT 10, Chloride titrator). Gills of fish used for bloodsampling were checked for mucus clogging. Water was sampled from fish tanks on 11 occasions. pH was measured immediately (ORION MOD.207 with a Radiometer glass-electrode) and aliquots were prepared for aluminium analyses. All analytical methods were identical to those described by SKOGHEIM and ROSSELAND (1984). Further details on the experimental conditions are given by ROSSELAND and SKOGHEIM (1982).

Calculation of aluminium speciation

Aluminium speciation was calculated using the computer program MINEQL (WESTALL *et al.* 1976), assuming chemical equilibrium. Inorganic monomeric species of aluminium were calculated from measurements of pH, fluoride, sulphate and labile aluminium. Ionic strength was estimated to be $6 \cdot 10^{-4}$ M. Several values for the solubility constant for:

$$Al(OH)_3 + 3H^+ = Al^{3+} + 3H_2O$$

were used. Equilibrium constants for the inorganic complexes used were similar to those noted by DRISCOLL (1980).

III. RESULTS

Water quality

Due to the acidity of the added $Al_2(SO_4)_3$, pH decreased from 4.99 in tank A to 4.90 in tank D (with the highest Al concentration). The pH of lake water (control) was constant at 6.6 during the experiment.

Aluminium in the experimental tanks occurred principally as labile (inorganic) forms. Concentrations of labile Al in each treatment were stable, with coefficients of variations < 5 % (Fig. 1). Concentrations of non-labile Al increased progressively from 56 μgAl/l in tank A to 69 μgAl/l in tank D (significant: t — test, $p < 0.01$) (Fig. 2 A). The concentration of organic matter (estimated as UV-absorbance) was stable and low during the experiment (ROSSELAND and SKOGHEIM 1982). Measured against a Pt-standard the level of humic material was about 5 mgPt/l. The chemical oxygen demand was of the order 1.0—1.5 mgO/l. Thus the chelating capacity of organic matter in the brook water did not influence aluminium speciation to any great extent. In the lake water no labile Al was detected as expected at that pH (6.6), the concentration of non-labile Al was 50 μgAl/l.

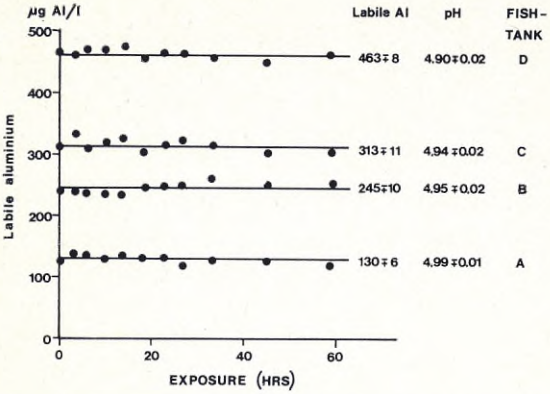


Fig. 1. Concentration of labile Al in the various fish tanks during the experiment. Mean values and standard deviation for 11 observations in each fish tank are given.

Estimated equilibrium concentrations of monomeric aluminium, assuming various solubility constants for $Al(OH)_3$, are illustrated in Fig. 2 B. Even slight variations in the solubility constant (or in temperature, given the influence of temperature on solubility) significantly alter the expected equilibrium concentration. WRIGHT and SKOGHEIM (1983) reported on aluminium speciation at the interface of an acid stream and limed lake. They concluded that the lake water at pH 5.5 was only slightly oversaturated with respect to synthetic gibbsite (log K = 9.3 at 5°C). Temperature in our experiment was 5.2°C. Assuming log K = 9.3 at 5.2°C, measured levels of labile Al were undersaturated relative to gibbsite in tank A, near equilibrium in tank B, and oversaturated in tanks

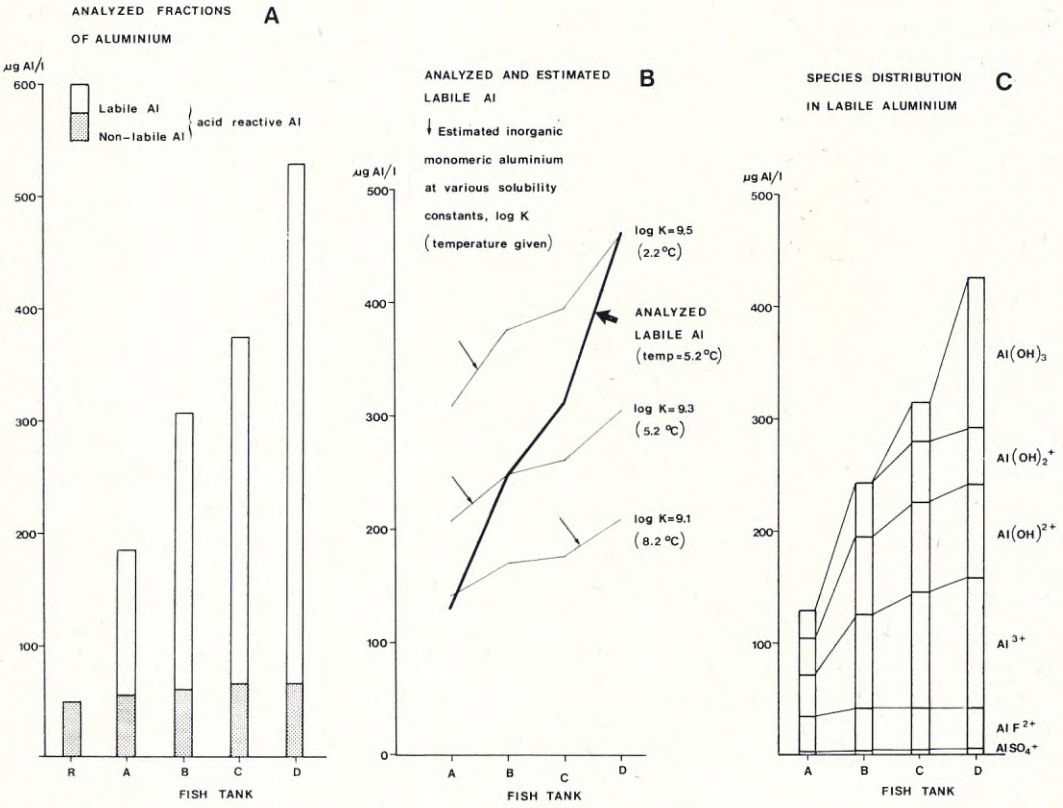


Fig. 2. A. Concentration of the analysed fractions of aluminium. B. Estimated concentrations of inorganic monomeric aluminium calculated at various solubility constants (log K) and the analysed concentration of labile Al. C. Calculated species distribution in the measured labile Al using solubility constant corresponding to that of synthetic gibbsite at the actual temperature (5.2°C).

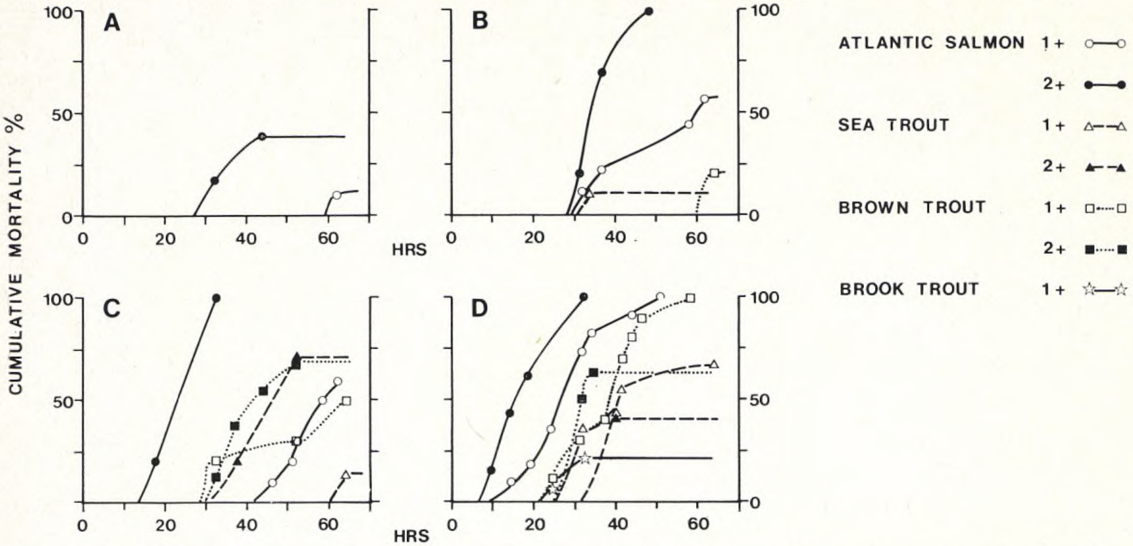


Fig. 3. Cumulative mortality of the different species and year classes in the four water qualities (A—D). Number of fish in each group varied between 8 and 14.

C and D (Fig. 2 B). This apparent oversaturation of aluminium in tank C and D, is supported by an incomplete recovery of added aluminium (40 and 70 µg Al/l, respectively), suggesting precipitation of aluminium out of solution.

Calculations of the major inorganic aluminium species based on measured labile Al and assuming log K=9.3 are illustrated in Fig. 2 C. Aquo Al^{3+} and aluminium-hydroxides species $Al(OH)^{2+}$ and $Al(OH)_2{}^+$ were the dominant forms in all tanks. Concentration of these species increased with increasing aluminium additions. The level of fluoride-complexed aluminium (ALF^{2+}) was almost constant and controlled by the constant concentration of fluoride (35 µgF/l). Aluminium-sulphate ($AlSO_4{}^+$) concentrations were very low.

The concentration of calcium in lake water was 4.2 mgCa/l; in brook water 2.0 mgCa/l. Alkalinity of lake water was 0.09 meq/l, but < 0 meq/l in brook water.

Mortality

Mortalities of the different species and year classes exposed to the four types of waters are illustrated in Fig. 3. Presmolt of salmon (age 2+) were the most sensitive, with 100 % mortality within 48 hrs at a mean concentration of labile Al of 245 µgAl/l (tank B). One-year-old salmon were less sensitive than two-year-old salmon, showing 100 % mortality within 64 hrs only in tank D at a mean labile Al concentration of 463 µgAl/l, but in general more sensitive than the other species tested. For the two forms of trout, sea trout and brown trout, two-year-old fish (presmolt of sea trout) were more sensitive than one-year-old fish only in tank C. Also in this treatment (but not in A, B or D), age 2+ sea trout and brown trout had a higher mortality than one-year-old salmon. In general, there seemed to be no systematic tendency for greater sensitivity of age 2+ for sea trout and brown trout. Brook trout had the lowest mortality in all treatments, with fish dying only at the highest concentration of labile Al (463 µgAl/l).

Prior to death, each fish went through a typical change in behaviour. Starting with reacting sluggishly in response to visual and sound stimuli, followed by abnormal swimming with periodes of lateral rest on bottom. Several hours before respiratory arrest, the fish lay motionless, apparently unable to move.

Plasma chloride

No mortality or loss of plasma chloride occurred in lake water. During the experiment, mean levels and standard deviation of plasma chloride (in meq/l) for fish in lake water were: salmon 124+5

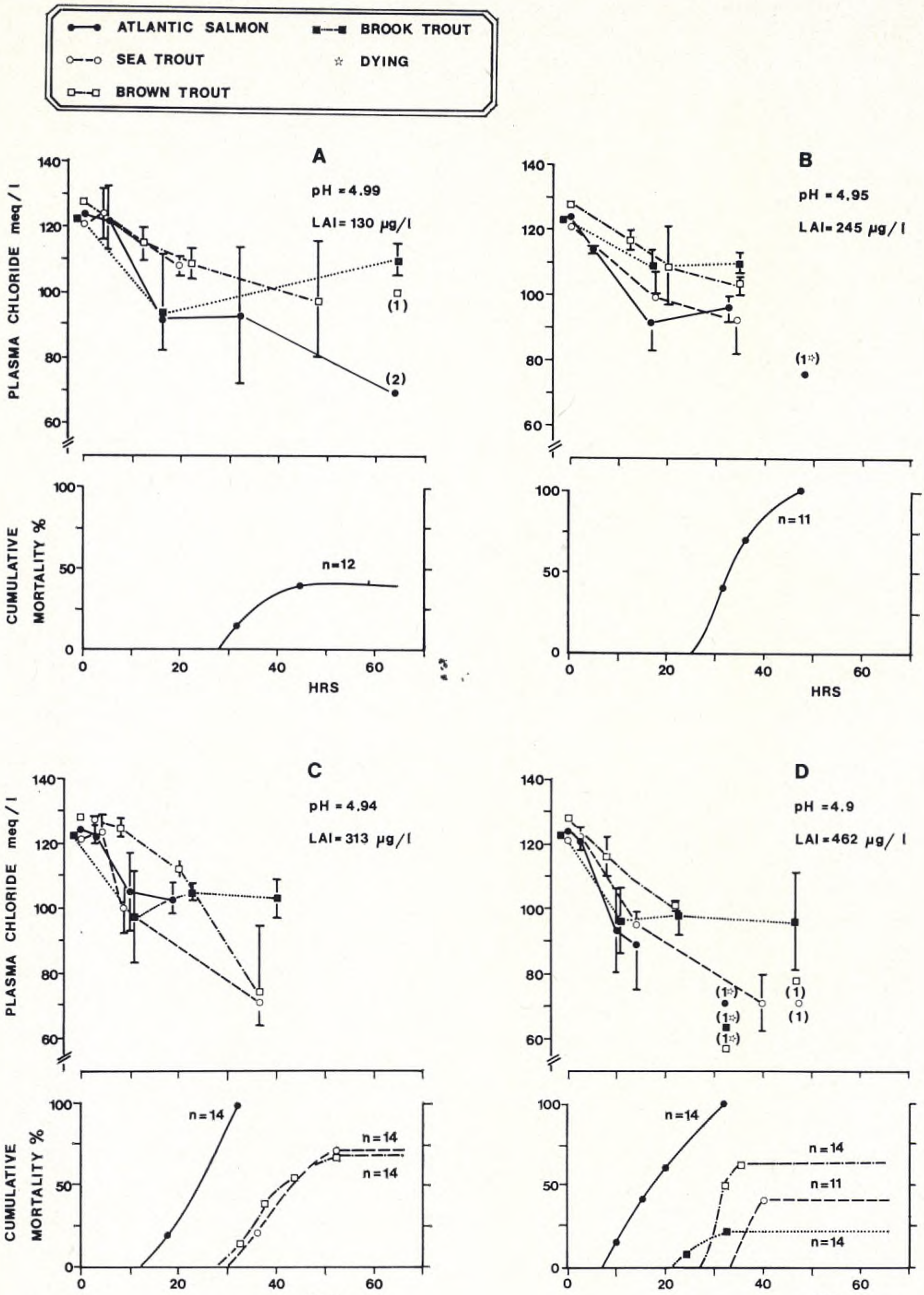


Fig. 4. Plasma chloride as a function of time in the different water qualities (A—D). Salmon, sea trout and brown trout were two-year old, brook trout one-year old. Mean and standard deviation (n=3) are given if not otherwise stated. For comparison, the cumulative mortality of the respective year classes, with the number of fish in each group (n), are given.

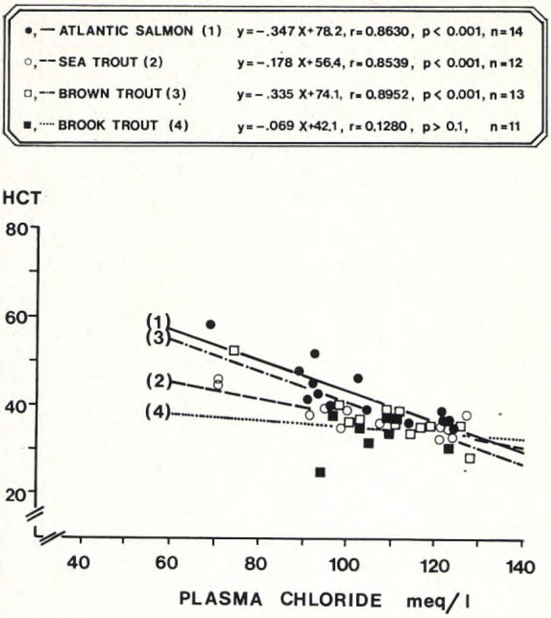


Fig. 5. The relation between mean values of hematocrit (HCT) and plasma chloride in the acid water qualities (A—D, pooled samples). The regression lines with the respective data are given.

(n=11), sea trout 121+4 (n=6), brown trout 128+3 (n=6) and brook trout 123+5 (n=6).

Changes in concentrations of plasma chloride as a function of time for two-year-old salmon, sea trout and brown trout and one-year-old brook trout in the different treatment waters, are shown in Fig. 4. Rate of ion loss generally reflects relative species sensitivity as measured by cumulative mortality. The initial loss rate was highest for salmon, with the loss rate increasing significantly ($p < 0.01$) with increasing levels of aluminium, from 0.88 meqCl/hr in treatment A, to 0.93 in B, 1.25 in C and 2.1 meqCl/hr in treatment D (ROSSELAND and SKOGHEIM 1982).

Although the mortality of sea trout and brown trout was similar, the initial loss of plasma chloride was more rapid for sea trout than brown trout in the higher aluminium concentrations (B—D). Brook trout response is noteworthy. Except in tank B, plasma chloride losses during the first 10—20 hrs for brook trout were similar to those for salmon. After the initial losses, however, concentrations of plasma chloride in brook trout stabilized, and in most cases increased (Fig. 4, A—C), while levels for salmon continued to decline. Except for salmon in tank B, between time 18 hrs and 32 hrs, no similar stabilization or increase in plasma chloride levels were observed for other species.

Hematocrit

No changes in hematocrit occurred in fish held in lake water during the experiment. Mean concentrations and standard deviations of hematocrit for the different species were: salmon 35+3 (n=11), sea trout 33+2 (n=6), brown trout 29+2 (n=6) and brook trout 31+3 (n=6). The relationship between mean values of plasma chloride and hematocrit for the different species in the four treatment waters (A—D, pooled samples), is presented in Fig. 5. With a decrease in plasma chloride, salmon, sea trout and brown trout all demonstrated a significant increase in hematocrit ($p < 0.001$). For brook trout, plasma chloride and hematocrit were not significantly correlated.

Mucus

No change in mucus could be seen on gills of fish kept in the natural brook water (tank A) compared to gills from fish kept in lake water (control), not even on gills of fish dying from the osmotic stress (salmon). Whenever aluminium (as $Al_2(SO_4)_3$) was added (tank B—D), excessive mucus (yellowish colour) appeared between the gill lamellae. For all species, mucus clogging increased with an increased addition of aluminium, independent of plasma chloride levels in the individual fish. When observed through light microscope, no signs of damage of the secondary lamellae were seen.

IV. DISCUSSION

Waters used in these experiments provided realistic values of pH and aluminium occurring during chronic and episodic acidification causing fish kills in nature (LEIVESTAD and MUNIZ 1976, SKOGHEIM *et al.* 1984). The concentration of labile aluminium varied between 130 and 463 μgAl/l and theoretical calculations indicated situations of undersaturation, near equilibrium and oversaturation of labile aluminium with respect to a solid phase of $Al(OH)_3$. pH and aluminium levels were similar to those measured during an episodic fish kill of spawning migrating Atlantic salmon in River Ogna (Skogheim *et al.* 1984).

The lower concentration of calcium in the brook water compared to the lake water (2.0 and 4.2 mgCa/l respectively) is unlikely to have caused any physiological problems *per se*, at least for brown trout, as water of pH 5, calcium of 0.4 mgCa/l and total aluminium of 30 μgAl/l (labile Al not measured) did not cause any loss of plasma ions in brown trout (Leivestad *et al.* 1980).

In our experiments, fish response to the different water qualities appeared to be concentration-, species- and age-dependent. In general, higher concentrations of aluminium resulted in increased ion loss and mortality. Calculation of aluminium speciation indicated that concentrations of the aluminium-hydroxides ($Al(OH)_2^+$ and $Al(OH)_3$ (s)) together with aquo Al^{3+} increased with increasing level of labile aluminium. Oversaturated aluminium solutions at pH levels 5.2 to 5.4 have been found to be particularly toxic in previous studies (Baker and Schofield 1980, 1982). In our experiment, however, it cannot be determined if the higher toxicity in treatments C and D is associated with oversaturation of aluminium *per se* or with high levels of labile Al in general.

Our results are in accordance with earlier studies indicating that Atlantic salmon are more sensitive than brown trout and sea trout to acid water (Jensen and Snekvik 1972); brook trout was the most tolerant (Grande *et al.* 1978). These data support the observation that Atlantic salmon was the first species to disappear from acidified rivers in Norway (Jensen and Snekvik 1972, Leivestad *et al.* 1976).

The youngest life stages are generally considered most sensitive to acid water, leading to recruitment failure and absence of younger year classes (Jensen and Snekvik 1972, Leivestad *et al.* 1976, Rosseland *et al.* 1980). Toxicological experiments with different year-classes, yolksac fry and yearlings of salmon (referred in Muniz 1981) and one- and two-year-old brook trout (Muniz and Grande 1974), have confirmed this.

Our results with salmon contradict these generalizations. Two-year-old (presmolt) salmon were more sensitive than one-year-old salmon. The smoltification period therefore seems critical. Saunders *et al.* (1983) also found that low pH during smoltification period increased mortality of salmon and disturbed the smoltification process, thereby reducing tolerance to high salinity waters. These factors make this species and life stage extremely vulnerable to acidification.

In contrast, presmolt (age 2+) of sea trout were not consistently more sensitive to acid waters than one-year-old sea trout (Fig. 3 C, D). The greater tolerance of sea trout smolts compared to salmon agrees with most field observations (Jensen and Snekvik 1972), but is contrary to observations from a fish kill in River Vikedal where differences in the relative mortality of the two species were small (Hesthagen and Skogheim 1984). Likewise few of the younger year classes of salmon and trout died during this incidence, confirming our experimental results.

Levels of plasma chloride in salmon presmolt declined rapidly with exposure to acid waters, with the loss rate increasing with increasing aluminium concentrations. Sea trout and brown trout also experienced declines in plasma chloride but at a slower rate. Ion losses were more rapid for sea trout than brown trout. Brook trout experienced an initial rapid loss of chloride, in most cases comparable to the salmon (Fig. 4, A, C, D). However, while other species continued to lose ions, eventually resulting in death, ion losses stabilized for brook trout, and in some cases plasma chloride concentrations even tended to increase (Fig. 4, A, B). These shifts in rate of ion loss occurred after 10—20 hrs at all concentrations of aluminium. McWilliams (1980 a) demonstrated a similar change in rate of sodium (Na) uptake in brown trout during sublethal pH-stress, from an initial rapid decrease in Na-uptake, to an increase after 10—20 hrs.

For the salmon, sea trout and brown trout, decreased plasma chloride was accompanied by increased hematocrit. Similar results have occurred in several experiments with salmonid fishes, *e.g.* Neville (1979), McDonald *et al.* (1980). According to Wood and McDonald (1982) the increase in hematocrit may result from a combination of three factors: reduced plasma volume, red blood cell (RBC) swelling and RBC mobilization. The relative importance of these three factors were not evaluated in this experiment.

For brook trout, hematocrit and plasma chloride were not significantly correlated. It is not known whether this difference in response reflects true variations among species in effects of acidity. McWilliams (1980 b) noted that for brown trout, hematocrit decreased with a reduction in plasma ions, in contrast to our results.

The absence of excessive mucus on gills of fish exposed (and dying) in the acid brook water with naturally occurring toxic aluminium concentrations (tank A), is in accordance with observations of gills of spawning migrating Atlantic salmon dying during an episodic fish kill caused by acid aluminium-rich water (Skogheim *et al.* 1984). Whenever aluminium was added (as $Al_2(SO_4)_3$) to the test waters, excessive mucus appeared on gills of each species. These observations are in accordance with other experiments where $Al_2(SO_4)_3$ have been added to the test waters (Muniz and Leivestad 1980, Rosseland 1980).

The mucus clogging observed in our and other experiments, might thus have been an effect of the experimental conditions. The ecological relevance of respiratory failure (due to reduced O_2 diffusion over the gill surface leading to hypoxia) as a cause of death in acidic (Ultsch and Gros 1979) and aluminium rich waters (Muniz and Leivestad 1980), is thus questioned.

V. ACKNOWLEDGMENT

We gratefully acknowledge the assistance given by the staff at the Research Station for Freshwaterfish, Ims, and the help from Jan Ruud Hansen in execution of the computer program.

A special thank to the Drs. Joan Baker, Magne Grande, Arne Henriksen, Kjell W. Jensen and Helge Leivestad for valuable comments and improvements of the manuscript.

VI. REFERENCES

Baker, J. P. 1982. Effects on fish of metals associated with acidification. p. 165—176. *In* Acid rain/fisheries. Ed.: R. E. Johnson. Proc. Int. Symp. Acidic Rain and Fishery Impacts on Northeastern North America. Am. Fish. Soc., Bethesda, Maryland.

— and C. L. Schofield. 1980. Aluminium toxicity to fish as related to acid precipitation in Adirondack surface water quality. p. 292—293. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

— and C. L. Schofield. 1982. Aluminium toxicity to fish in acidic waters. Wat., Air, Soil. Poll. 19: 289—309.

Brown, D. J. A. 1983. Effects of calcium and aluminium concentration on the survival of brown trout (*Salmo trutta*) at low pH. Bull. Environ. Contam. Toxicol. 30: 582—587.

Bua, B. and E. Snekvik. 1972. Klekkeforsøk med rogn av laksefisk 1966—1971. Virkning av surhet og saltinnhold i klekkevannet. (Hatching experiments with roe from salmonid fishes 1966—1971. Effects of acidity and dissolved salt in the water.) Vann 7: 86—93. (In Norwegian.)

Driscoll, C. T. 1980. Chemical characterization of some dilute acidified lakes and streams in the Adirondack region of New York State. Ph. D. Thesis, Cornell Univ., Ithaca, N.Y. 308 p.

— J. P. Baker, J. J. Bisogni Jr. and C. L. Schofield. 1980. Effect of aluminium speciation on fish in dilute acidified water. Nature, Lond. 284: 161—164.

Grande, M., I. P. Muniz and S. Andersen. 1978. The relative tolerance of some salmonids to acid waters. Verh. Int. Verein. Limnol. 20: 2076—2084.

Hesthagen, T. and O. K. Skogheim. 1984. High mortality of presmolt of Atlantic salmon, *Salmo salar* L. and sea trout, *Salmo trutta* L. during spring snow melt 1982 in River Vikedalselva, Western Norway. (Manuscript.)

Huitfeldt-Kaas, H. 1922. Om aarsaken til massedød av laks og ørret i Frafjordelven, Helleelven og Dirdalselven i Ryfylke høsten 1920. (On the cause of mass death of salmon and trout in Frafjordelven, Helleelven and Dirdalselven in Ryfylke, autumn 1920). Norsk Jæger- Fiskefor., Tidsskr. 1/2: 37—44. (In Norwegian.)

Jensen, K. W. and E. Snekvik. 1972. Low pH levels wipe out salmon and trout populations in southern Norway. Ambio 1: 223—225.

Johansson, N., P. Runn and G. Milbrink. 1977. Early development of three salmonid species in acidified water. Zoon, Uppsala 5: 127—132.

Leivestad, H. and I. P. Muniz. 1976. Fish kill at low pH in a Norwegian river. Nature, Lond. 259: 391—392.

— G. Hendrey, I. P. Muniz and E. Snekvik. 1976. Effects of acid precipitation on freshwater organisms. p. 87—111. *In* Impact of acid precipitation on forest and freshwater ecosystem in Norway. Ed.: F. H. Brække. SNSF-project FR 6/76. Oslo—Ås.

— I. P. Muniz and B. O. Rosseland. 1980. Acid stress in trout from a dilute mountain stream. p. 318—319. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

McDonald, D. G., H. Hobe and C. M. Wood. 1980. The influence of calcium on the physiological responses of the rainbow trout, *Salmo gairdneri*, to low environmental pH. J. exp. Biol. 88: 109—131.

McWilliams, P. G. 1980 a. Effects of pH on sodium uptake in Norwegian brown trout (*Salmo trutta*) from an acid river. J. exp. Biol. 88: 259—267.

— 1980 b. Acclimation to an acid medium in the brown trout *Salmo trutta*. J. exp. Biol. 88: 269—280.

Muniz, I. P. 1981. Acidification and the Norwegian salmon. *In:* Acid rain and The Atlantic salmon. Int. Atlantic Salmon Found. Spec. Publ. 10: 65—72.

— and M. Grande. 1974. Overleving av ulike arter laksefisk i vann fra et surt vassdrag. (Survival of various species of salmonid fish in water from an acid water-course.) p. 29—39. *In* Hydrokjemiske og hydrologiske rapporter fra NIVA. Ed.: F. H. Brække. SNSF-project IR 3/74. Oslo—Ås. (In Norwegian.)

— and H. Leivestad. 1980. Toxic effects of aluminium on the brown trout, *Salmo trutta* L. p. 320—321. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

Neville, C. M. 1979. Sublethal effects of environmental acidification on rainbow trout (*Salmo gairdneri*). J. Fish. Res. Bd. Can. 36: 84—87.

Rosseland, B. O. 1980. Effects of acid water on metabolism and gill ventilation in brown trout, *Salmo trutta* L., and brook trout, *Salvelinus fontinalis* Mitchill. p. 348—349. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

— I. Sevaldrud, D. Svalastog and I. P. Muniz. 1980. Studies of freshwater fish populations — effects of acidification on reproduction, population structure, growth and food selection. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

— and O. K. Skogheim. 1982. Physiological stress and mortality of Atlantic salmon, *Salmo salar* L. in acid water with high levels of aluminium. ICES, C.M. 1982/M: 29. 15 p.

Rosseland, L. 1953. Om virksomheten i 1948. (Annual Report 1948. Report from the Fishery Inspector to the Department of Agriculture for the years 1948—50.) Oslo, Norway. (In Norwegian.)

Saunders, R. L., E. B. Henderson, P. R. Harman, C. E. Johnston and J. G. Eales. 1983. Effects of low environmental pH on smolting of Atlantic salmon (*Salmo salar*). Can. J. Fish. Aquat. Sci. 40: 1203—1211.

Schofield, C. L. 1977. Acid snow-melt effects on water quality and fish survival in the Adirondack Mountains of New York State. Res. Proj. Tech. Comp. Rep. Project A-072-NY. Cornell Univ. Ithaca, New York. 27 p.

Skogheim, O. K. and B. O. Rosseland. 1984. A comparative study on salmonid fish species in acid aluminium-rich water. I. Mortality of eggs and alevins. Rep. Inst. Freshw. Res., Drottningholm 61: 177—185.

Skogheim, O. K., B. O. Rosseland and I. Sevaldrud. 1984. Deaths of spawners of Atlantic salmon in River Ogna, SW Norway, caused by acidified aluminium-rich water. Rep. Inst. Freshw. Res., Drottningholm 61: 195—202.

Ultsch, G. R. and G. Gros. 1979. Mucus as a diffusion barrier to oxygen: Possible role in O_2 uptake at low pH in carp (*Cyprinus carpio*) gills. Comp. Biochem. Physiol. 62 A: 685—689.

Westall, J. C., J. L. Zacjary and F. N. M. Morel. 1976. MINEQL, a computer program for the calculation of chemical equilibrium composition of aqueous systems, Dep. Civil Eng. MIT, Tech. Note 18. 91 p.

Wood, C. M. and D. G. McDonald. 1982. Physiological mechanisms of acid toxicity to fish. p. 197—226. *In* Acid rain/fisheries. Ed.: R. E. Johnson. Proc. Int. Symp. Acidic Rain and Fishery Impacts on Northeastern North America. Am. Fish. Soc., Bethesda, Maryland.

Wright, R. F. and O. K. Skogheim. 1983. Aluminium speciation at the interface of an acid stream and a limed lake. Vatten 39: 301—304.

Deaths of Spawners of Atlantic Salmon (*Salmo salar* L.) in River Ogna, SW Norway, Caused by Acidified Aluminium-Rich Water

ODD K. SKOGHEIM, BJØRN OLAV ROSSELAND and IVER H. SEVALDRUD

Directorate for Wildlife and Freshwater Fish, Fish Research Division, P. O. Box 63, N-1432 Ås-NLH, Norway

ABSTRACT

Three kilometres upstream the outlet of River Ogna, very acid aluminium-rich water (pH=4.7, labile Al=345—360 µg/l) from a hydroelectric power plant is discharged into the main river. During a period in August 1982 the water in the main river was of good quality (pH=6.0, labile Al=13 µg/l). On increasing water flow in the main river the spawners of Atlantic salmon (*Salmo salar* L.) migrated from the estuary into the river. Then the water flow in the main river decreased and the acid aluminium-rich water from the hydroelectric power plant strongly influenced the water quality (pH=5.17—5.54, labile Al=109—133 µg/l). During a period of ca 35 hours more than 50 fish with weights 3 to 10 kg were found dead or dying in the river. Bloodsamples from dying salmon showed mean plasma-chloride levels of 110 meq/l, indicating significant loss of plasma-ions.

This episode indicates high sensitivity of migrating spawners of salmon to acid water oversaturated with aluminium.

I. INTRODUCTION

Acidification threatens all life stages in freshwater of salmonid fish species. In general recruitment failure due to high mortality of the youngest stages (egg-fry-fingerlings) have been considered as the main cause for fish disappearence (JENSEN and SNEKVIK 1972, ROSSELAND *et al.* 1980). In some few cases fish-kill involving older fish have been reported (HUITFELDT-KAAS 1922, DAHL 1927, ROSSELAND 1953, LEIVESTAD and MUNIZ 1976). High mortality of postspawners of brown trout in an acidified river system have also been reported (ROSSELAND *et al.* 1980). Both in laboratory experiments (ROSSELAND and SKOGHEIM 1982, 1984) and in nature (HESTHAGEN and SKOGHEIM 1984 MS) high mortality of presmolt/smolt of Atlantic salmon (*Salmo salar* L.) in acid, aluminium-rich water have been found.

An episodic kill of migrating spawners of Atlantic salmon in a river in SW Norway was observed as early as in 1920 (HUITFELDT-KAAS 1922). No convincing explanation of the cause was given. In the nearby River Ogna an episodic mortality of migrating spawners of Atlantic salmon was observed in August 1982. In this paper this fish-kill will be discussed in relation to water quality.

II. THE STUDY AREA

River Ogna is situated in SW Norway in the county of Rogaland. The bedrock is largely composed of plagioclase andesine, called the Egersund-anorthosite. The elevated parts of the area are poor in vegetation and quaternary deposits. In the main valley the agricultural activity is rather intense. The total catchment area is 112 km^2 and 42 km^2 of this represents catchments being transferred to River Ogna for hydroelectric power production. In Fig. 1 is shown the lower part of the river and the outlet from the hydroelectric power station is marked out (site IV). The average waterflow is 4.1 m^3/s at site V.

The area has a temperate continental climate with mild winters and moderate summers. The annual precipitation amounts to ca 2000 mm (NORWEGIAN METEROLOGICAL INSTITUTE 1972). The wet-deposition of sulphur (corrected for sea-sulphate) amounts to ca 1.2 gS/m^2 (STATE POLLUTION CONTROL AUTHORITY 1982). Because of relatively small water reservoirs in the catchment, the waterflow in the main river fluctuates with changes in precipitation. A considerable fluctuation in pH occurs with generally low pH during the winter and higher pH during summer (Fig. 2). The water from the hydroelectric power station is cronically acid, pH 4.7.

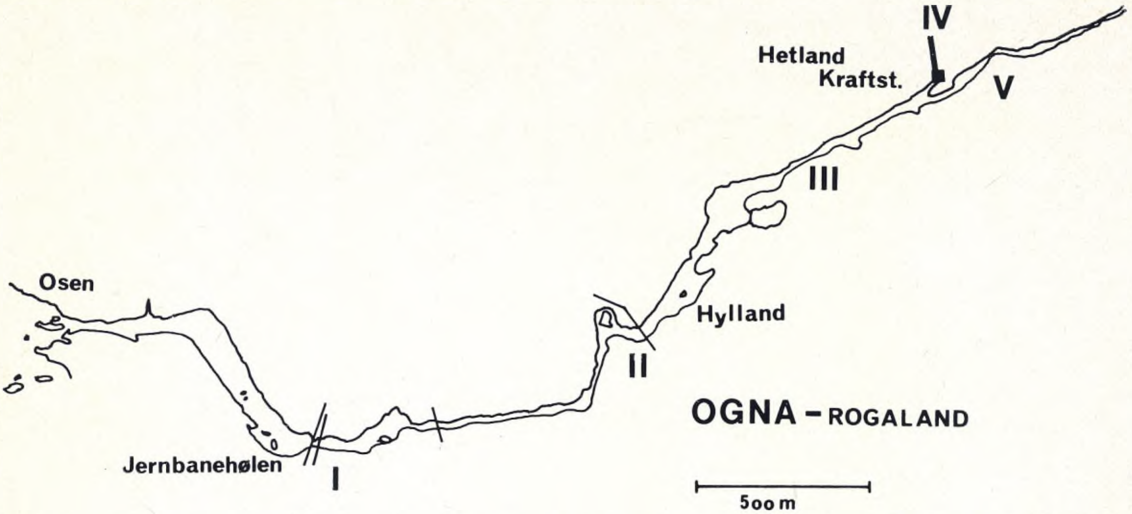


Fig. 1. Location of sampling sites in River Ogna, SW Norway. The map shows the sampling sites in the main river (V), upstream the outlet (IV) of Hetland Hydroelectric Power Station and downstream (I, II and III) on the river stretch to the estuary.

Fish populations in the river are Atlantic salmon (*Salmo salar* L.), brown trout and sea trout (*Salmo trutta* L.), and eel (*Anguilla anguilla* L.). Salmon is the most popular fish for angling and the catch varied between 152 and 4180 kg/year during the last 15 years.

In September 1969 there was a fish-kill of fingerlings and adult salmon and trout. This event took place during heavy autumn rains after a long period of dry weather (SNEKVIK 1975). During the winter 1982 an extensive fish-kill of salmon occurred (HESTHAGEN *et al.* 1982). This coincided with the very low pH measured in that period (Fig. 2).

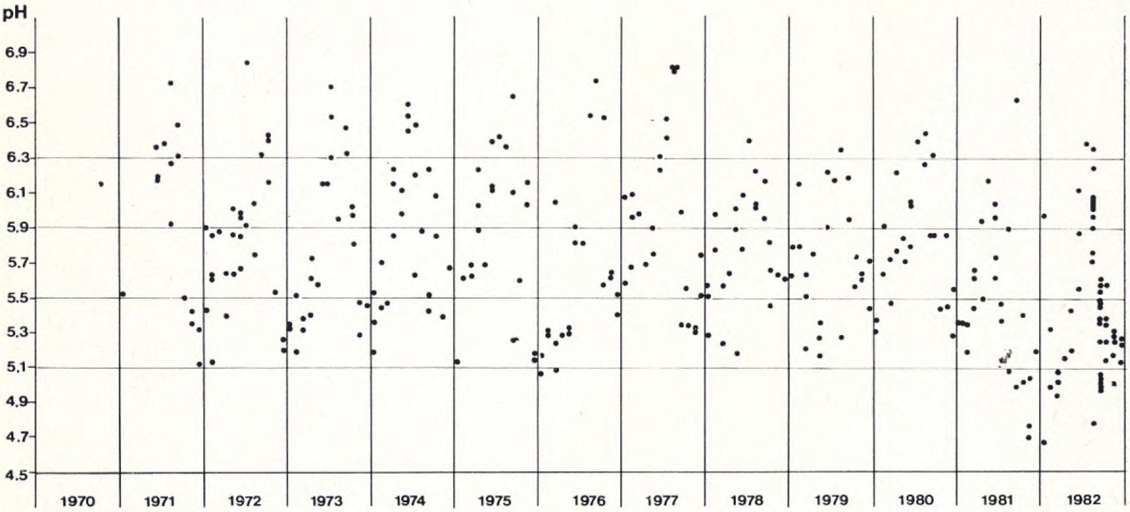


Fig. 2. pH observations in River Ogna (sampling site V, Fig. 1) during the last decade.

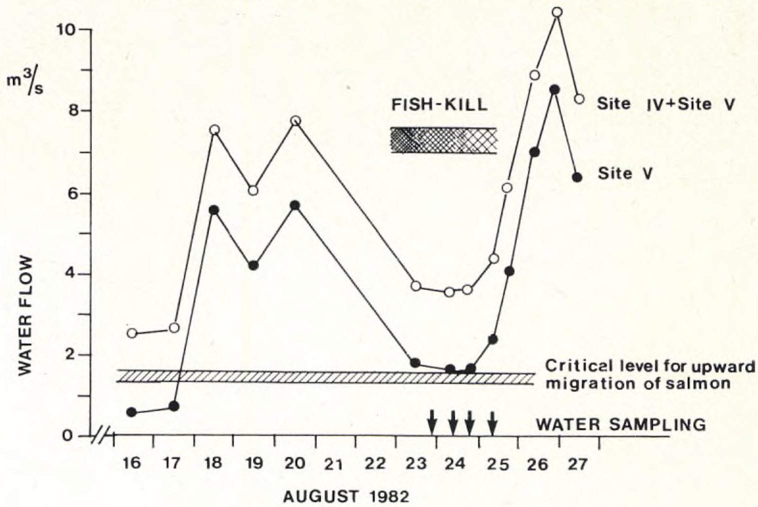


Fig. 3. Waterflow in River Ogna, SW Norway. Location of sampling site V in the main river upstream the outlet of Hetland Hydroelectric Power Station (IV) — (Fig. 1). The period of fish kill, time of water sampling and the critical level for upstream migration of Atlantic salmon are marked in the figure.

III. MATERIAL AND METHODS

Water quality

Water samples were collected at sites I—V at several occasions during the episode (Fig. 3). In the field samples were prepared for determination of three fractions of aluminium: total Al, acid-reactive Al and non-labile Al. The methods are slightly modified after DRISCOLL (1980). Aluminium was analysed by the pyrocatechol violet method (DOUGAN and WILSON 1974). In order to minimize contamination and handling the analyses were performed directly in the sample tubes containing aliquots prepared in the field. Total Al was analysed after digestion with $K_2S_2O_8$. Acid-reactive Al was analysed directly without any pretreatment. Samples passed through a cation-exchange column (DOWEX -50, X8, 50/100 mesh prepared with Na^+) was analysed as acid-reactive Al and was assumed to mainly include organic complexed non-labile Al. The difference between acid-reactive Al and non-labile Al fraction is called labile monomeric Al and is assumed to include inorganic fractions as free aluminium and soluble aluminium hydroxide, flouride and sulphate complexes. pH was measured with a Radiometer pHM 80. Calcium and magnesium was determined by atomic absorption spectrophotometry (Perkin—Elmer Mod. 603). Alkalinity was determined by automatic titration with a Radiometer RTS 822. Dissolved organic matter was estimated by UV-absorbance at 254 nm, and by a Pt-comparator. Sulphate and chloride was analysed by ion-exchange technique in combination with conductometry (modified after STAINTON 1974). Total fluoride was analysed using an ion-selective electrode (ORION 1976). Water flow in the main river (site V) was registered at the monitoring station of the Norwegian Electricity Board.

Fish observations

The river stretch from site I to V was surveyed for dead and dying fish several times during the episode. At site I three salmon with irregular behaviour were caught by landing net and blood-samples were drawn directly by heart puncture. The blood was immediately centrifuged (MSE micro hematocrit centrifuge type 346, 3 min.) and plasma samples were analysed for chloride (Radiometer CMT 10 Chloride titrator). The fish gills were cut off from nine fish (including the three used for blood samples). Duplicate fractions of the gill arches (0.2—1.0 g fresh weight) were digested in 1 N HCl. The supernatant was autoclaved with $K_2S_2O_8$ and Al was determined by atomic absorption spectrophotometer equipped with graphite furnace.

Calculation of aluminium speciation

Aluminium speciation was calculated by the computer program MINEQL (WESTALL *et al.* 1976).

The dissolved species of aluminium were assumed to be in chemical equilibrium. Free (aquo) aluminium was calculated with measured pH, fluoride, sulphate and the measured labile, monomeric aluminium. Using this value for aquo aluminium, the theoretical coexisting dissolved aluminium complexes with hydroxide, fluoride and sulphate were calculated. For the equilibrium between aquo aluminium and aluminiumtrihydroxide

$$Al(OH)_3 + 3H^+ = Al^{3+} + 3H_2O$$

a solubility constant corresponding to that of synthetic gibbsite was used (MAY *et al.* 1979). SEIP *et al.* (1984) showed the importance of temperature when selecting solubility constants of gibbsite. The log K value of synthetic gibbsite is 8.11 at 25°C. This value was converted to *in situ* temperature (site IV: 10°C—log K 9.0, site I—II—III: 12°C log K 8.8). The thermodynamic equilibrium constants for the inorganic complexes used were practically the same as those used by DRISCOLL (1980). The ion-strength was estimated to $7 \cdot 10^{-4}$M.

IV. RESULTS

Variations in water flow during the period of interest to the fish-kill is shown in Fig. 3. The discharge from the hydroelectric power station was constant at 1.9 m³/s while the flow in the main river varied considerably. The water level needed for the spawners of salmon to migrate from the estuary into the river is marked in Fig. 3. The first five days after August 17 there were good fishing results according to the anglers. In the evening of August 22 the first dead or dying fish were observed. Along the river stretch between sites I—III (Fig. 1) dead fish could be seen lying on the bottom of the river or floating downstream. Dying fish could be observed due to abnormal behaviour. During a period of 2—3 days (Fig. 3) at least 50 dead or dying specimens of salmon were observed. Most fish were infected by the sea lice (*Lepeophteirus* spp.) and were therefore spawners recently migrated from the estuary. The size of the fish varied between 3 and 10 kg.

At site I three salmon with abnormal behaviour were caught by landing net and analysed for plasma chloride. The concentrations of plasma chloride were 103, 107 and 113 meq/l, respec-

Table 1. *Chemical composition of some selected water samples taken during the fish-kill episode in River Ogna. Sampling sites refer to Fig. 1. In the table are listed values of pH, calcium (Ca), magnesium (Mg), sulphate (SO₄), chloride (Cl), alkalinity (Alk), total fluoride (F), total aluminium, acid-reactive aluminium, non-labile aluminium and dissolved organic matter (DOM) estimated as ultraviolet absorbance.*

Date	Sampling site	Time hour	pH	Ca mg/l	Mg mg/l	SO_4 mg/l	Cl mg/l	ALK μeq/l	F μg/l	Total Al μg/l	Acid reactive Al μg/l	Non-labile Al μg/l	DOM $\varepsilon^{254\ nm}_{1\ cm}$
Aug. 23, 1982	I	22	5.54	2.02	1.15	6.6	14.0	5	22	164	155	25	0.027
„ 23, „	IV	22	4.68	0.92	1.00	6.2	13.8	0	21	371	370	7	0.008
„ 24, „	I	9	5.45	2.14	1.19	6.7	14.3	< 5	23	157	155	22	0.027
„ 24, „	IV	9	4.69	0.93	0.98	6.2	14.7	0	20	364	352	6	0.004
„ 24, „	V	9	6.18	2.20	1.16	6.2	13.0	18	22	47	43	30	0.051
„ 24, „	I	20	5.39	1.75	1.06	6.8	14.7	< 5	20	189	187	80	0.027
„ 25, „	I	9	5.41	1.66	1.03	6.7	14.7	< 5	21	198	181	72	0.027
„ 25, „	II	9	5.15	1.30	0.95	6.4	14.6	0	22	202	177	49	0.031
„ 25, „	III	10	4.76	0.96	0.99	6.6	14.4	0	21	362	352	45	0.012
„ 25, „	IV	11	4.69	0.87	0.96	6.5	14.4	0	22	394	375	30	0.004
„ 25, „	V	11	5.96	1.92	1.06	6.4	13.8	14	22	95	72	60	0.051

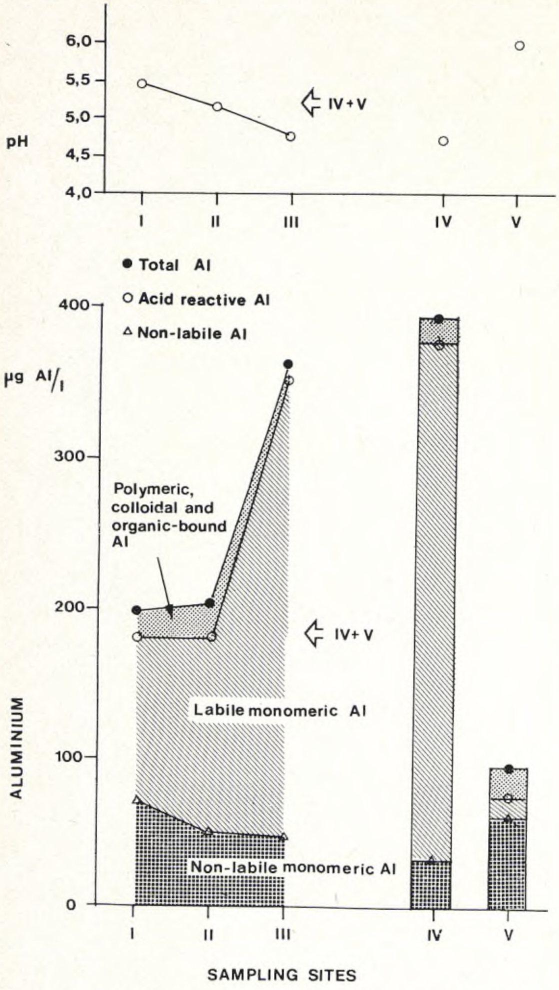


Fig. 4. pH and the measured fractions of aluminium in River Ogna at sampling date 820825.

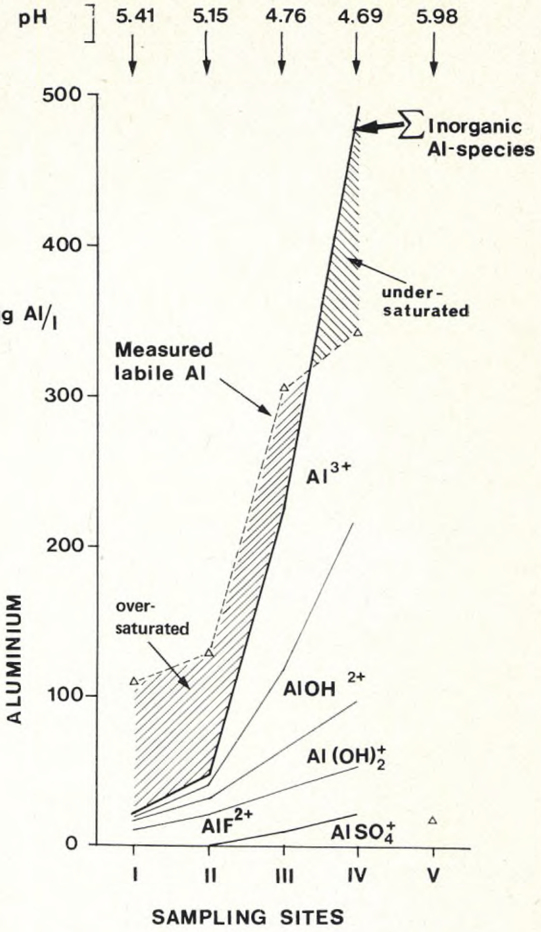


Fig. 5. Species of dissolved inorganic aluminium in River Ogna (date 820825, Fig. 4) computed by MINEQL. The equilibrium constant of synthetic gibbsite (log K=8.11 at 25°C) was applied after conversion to the *in situ* temperature. The fraction of labile Al measured as the difference between acid reactive Al and non-labile Al is plotted.

tively, and compared to the normal range 120—135 meq/l it is obvious that the osmoregulation of the fish was disturbed.

The concentration of aluminium on gills from dead or dying fish varied from 70—341 µg Al/g fresh weight with a mean value of 166 (n=9). Compared to values of fingerlings of salmon from River Vikedal (HESTHAGEN and SKOGHEIM 1984), which may be taken as reference, there was an enrichment of aluminium by an order of 10.

The water sampling during the episode of fish-kill is marked on Fig. 3. In Table 1 some of the chemical results are given, and in Fig. 4 and 5 the results from August 25 are treated in detail. The water from the outlet of the hydroelectric power station (site IV) is cronically acid with pH 4.7 and rich in aluminium. The main fraction is present as labile monomeric Al. The water is extremely low in dissolved and particulate organic matter and the concentration of non-labile Al (mainly organic-complexed) and polymeric forms of Al is therefore low (Fig. 4). The water quality in the main river at site V varies considerably throughout the year (Fig. 2), but during this episode the pH was around 6.0 and some buffer capacity, as alkalinity, was present. In this water the labile fraction of Al was very low (12 µg Al/l) and the dominant fraction was non-labile Al in agreement with the higher level of dissolved organic matter in this water. These two types of water qualities are mixed together and the resulting water quality at site I—III depends on the quantities, mixing and kinetics of chemical reactions.

At the sampling date shown in Fig. 4, pH rose from 4.7 at site IV to 5.5 at site I due to the buffering capacity of the water in the main river. After mixing of the two water qualities the most striking change appears in the speciation of aluminium. The speciation of aluminium will be dealt with in detail. This metal was the only one (among Fe, Mn, Cd, Zn, Pb, Cu) present in potentially toxic concentration.

The calculated inorganic species of aluminium present at the actual pH, sulphate and fluoride concentration is shown in Fig. 5. These species are added up and can be compared to the measured concentration of labile monomeric Al.

It appears that at site IV the water is sligthly undersaturated and at sites I—III the water is oversaturated with respect to synthetic gibbsite.

At the lowest pH (site IV) the dominant forms of aluminium is free Al^{3+} and aluminium hydroxide $AlOH^{2+}$. At higher pH (site I and II) only small concentrations of these species should be present and the high concentration of labile Al measured indicate oversaturation with respect to a solid phase of $Al(OH)_3$.

V. DISCUSSION

In the lower part of River Ogna two types of water were mixed together in variable quantities. Cronically acid (pH 4.7), aluminium-rich (350 µg/l of labile Al) water was discharged from a hydroelectric power station into the main river which at the time of the fish-kill was slightly buffered at pH 6.0. The mixing of these water qualities and the chemical reactions that occurred along the unidirectional movement of the water, thus created chemical gradients along the river stretch where the fish-kill appeared. On August 17 the water flow in the main river increased rapidly above the critical level for upward migration of salmon. For several days the water flow remained high enough to ensure de-acidification and intoxification of the water from the hydroelectric power station. When the fish-kill first was reported, on the evening of August 22, the water of the two types was mixed in equal proportions. During the fish-kill, which lasted for 2—3 days, the fish were trapped in the river. Some fish might, however, have escaped by downstream migration, a phenomenon observed by anglers from a bridge close to site I. Upstream migration was fatal because of the increasing gradient of acidity and concentration of labile aluminium towards the mixing site.

The water quality downstream the mixing-site depended both on the degree of mixing and the kinetics of the involved chemical reactions. At site I complete mixing is supposed to have been achieved. This appears from the fact that the concentration of total aluminium as well as conservative elements as calcium is in accordance with that estimated from simple mass law calculations. Due to the slow kinetics for precipitation of aluminium trihydroxides with a moderate pH-increase, the water at sites I—III appeared to be oversaturated with respect to synthetic gibbsite. This conclusion is of course dependent on the solubility constant used in the computation.

In this paper a solubility constant corresponding to that of synthetic gibbsite (log K=8.11 at 25°C (May *et al.* 1979)) is converted to the actual temperature. The converted solubility constant for synthetic gibbsite to the actual temperature (log K 8.8—9.0) was almost the same as that found by Driscoll (1980) and Johnson *et al.* (1981). From site III to I pH increased from 4.69 to 5.41 and the concentration of labile aluminium decreased from 345 µg/l to 109 µg/l. Driscoll *et al.* (1980) concluded that only inorganic forms of aluminium were toxic to postlarvae of fish and Baker and Schofield (1982) found that aluminium was most toxic in oversaturated solutions at pH levels 5.2 to 5.4. Higher sensitivity of Atlantic salmon during smoltification compared to earlier life stages was found by Rosseland and Skogheim (1982, 1984). In these experiments 50 % mortality occurred within 15—50 hours for presmolt of Atlantic salmon in the same aluminium range as in Ogna at pH 5.0. Smolts of salmon have also been shown to be the most sensitive compared to different year classes of other salmonid fish species (ROSSELAND and SKOGHEIM 1984).

Both during smoltification and during the preparation for the spawners to return from sea to freshwater, great physiological and hormonal changes occur in the fish. Although the sensitivity with respect to aluminium toxicity of spawners of Atlantic salmon just migrated from the estuary is not known from controlled experiments, high sensitivity is nevertheless likely. The presence of sea-lice on the killed spawners indicate their short stay in the river and high sensitivity of salmon at this stage can therefore be suggested.

Although gill damage (SCHOFIELD 1977) or mucus clogging of gills have been reported as a response to aluminium toxicity (MUNIZ and LEIVESTAD 1980, ROSSELAND 1980), this was not seen on the gills of the dying salmon during this fish kill. In the experiments on salmon presmolt (ROSSELAND and SKOGHEIM 1982, 1984) no mucus clogging was seen in the acid and toxic brook water, whereas addition of inorganic aluminium increased the mucus clogging of the gills. This should indicate that this phenomenon might occur only in situations with oversaturation and chemical disequilibrium of aluminium species. However, the situation in River Ogna did not impose mucus clogging in spite of oversaturation of aluminium with respect to a solid phase, but the concentration of aluminium on the gills increased by an order of 10. This is higher than found by DICKSON (1979) where the increased level of aluminium on gills of salmon fingerlings was in the order of 2—5 in toxic, acid, aluminium-enriched water at pH 5.0.

The fish-kill lasted for 2—3 days and only spawners of Atlantic salmon were involved. Given the same sensitivity of spawners as smolts of Atlantic salmon, a period of 2 days should be enough to reach a total mortality according to experiments with presmolt and smolt of Atlantic salmon (ROSSELAND and SKOGHEIM 1982, 1984). This may be the reason why the fish-kill ceased just prior to the increased water flow in the main river.

No other fish species or stages of salmon were involved in the fish-kill. Visual observations and electrofishing did only prove the presence of spawners of salmon. Other species and stages of salmon might therefore have been absent.

The situation described here with a discharge of acid, aluminium-rich water into a river with slightly buffered water is not unique. The situation is analogous to the interface between limed (or not acidified) and acid water (WRIGHT and SKOGHEIM 1983). Such systems should therefore be given great concern if valuable fish stocks are present downstream.

VI. ACKNOWLEDGMENT

We gratefully acknowledge the comments given by Dr. KJELL W. JENSEN and the valuable help with calculating aluminium speciation given by Dr. HANS MARTIN SEIP.

VII. REFERENCES

BAKER, J. D. and C. L. SCHOFIELD. 1982. Aluminium toxicity to fish in acidic waters. Wat. Air Soil Poll. 18: 289—309.

DAHL, K. 1927. The effect of acid water on trout fry. Salm. Trout Mag. 46: 35—43.

DICKSON, W. 1979. Exempel på metalltoxicitet vid försuring och kalkning. (Metaltoxicity in connection with acidification and liming.) Aquannalen, Univ., Lund 1: 2—7. (In Swedish.)

DOUGAN, W. K. and A. L. WILSON. 1974. The absorptiometric determination of aluminium in water. A comparison of some chromogenic reagents and the development of an improved method. Analyst, Camb. 99: 413—430.

DRISCOLL, C. T. 1980. Chemical characterization of some dilute acidified lakes and streams in the Adirondack Region of New York State. Ph. D. Thesis. Cornell Univ. 308 p.

— and J. P. Baker, J. J. BISOGNI JR and C. L. SCHOFIELD. 1980. Effects of aluminium speciation on fish in dilute acidified waters. Nature, Lond. 284: 161—164.

HESTHAGEN, T., I. SEVALDRUD and O. K. SKOGHEIM. 1982. Ogna- og Helgåvassdraget i Rogaland. Fiskestatus og vannkvalitet (River Ogna and River Helgå in County of Rogaland. Fish populations and water quality.) Dir. Wildl. Freshw. Fish., Rep. Fish Res. Div. 4. 34 p. (In Norwegian.)

— and O. K. SKOGHEIM. 1984. High mortality of presmolt of Atlantic salmon, *Salmo salar* L. and sea trout, *Salmo trutta* L. during spring snow melt 1982 in River Vikedalselva, Western Norway. (Manuscript.)

HUITFELDT-KAAS, H. 1922. Om aarsaken til massedød av laks og ørret i Frafjordelven, Helleelven og Dirdalselven i Ryfylke høsten 1920. (On the cause of mass death of salmon and trout in Frafjordeelven, Helleelven and Dirdalselven i Ryfylke, autumn 1920.) Norsk Jæger Fiskefor. Tidskr. 1/2: 37—44. (In Norwegian.)

JENSEN, K. W. and E. SNEKVIK. 1972. Low pH levels wipe out salmon and trout populations in southern Norway. Ambio 1: 223—225.

JOHNSON, N. M., C. T. DRISCOLL, J. S. EATON, G. T. LIKENS and W. H. MCDOWELL. 1981. "Acid rain", dissolved aluminium and chemical weathering at the Hubbard Brook Experimental Forest, New Hampshire. Geochim. Cosmochim. Acta 45: 1421—1437.

LEIVESTAD, H. and I. P. MUNIZ. 1976. Fish kill at low pH in a Norwegian river. Nature, Lond. 259: 391—392.

MAY, H. M., P. A. HELMKE and M. L. JACKSON. 1979. Gibbsite solubility and thermodynamic properties of hydroxy-aluminium ions in aqueous solution at 25°C. Geochim. Cosmochim. Acta 43: 861—868.

MUNIZ, I. P. and H. LEIVESTAD. 1980. Toxic effects of aluminium on the brown trout, *Salmo trutta* L. p. 320—321. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

NORWEGIAN METHEOROLOGICAL INSTITUTE. 1972. Precipitation measurements in Norway. Aschehoug & Co, Oslo. 104 p.

ORION. 1976. Instruction Manual. Fluoride Electrodes, Orion Research Inch., Cambridge.

ROSSELAND, B. O. 1980. Effects of acid water on metabolism and gill ventilation in brown trout, *Salmo trutta* L., and brook trout, *Salvelinus fontinalis* MITCHILL. p. 348—349. *In* Ecological impact of acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

— I. SEVALDRUD, D. SVALASTOG and I. P. MUNIZ. 1980. Studies on freshwater fish populations — effects of acidification on reproduction, population structure, growth and food selection. p. 336—337. *In* Ecological impact acid precipitation. Proc. Int. Conf., Sandefjord, Norway 1980. Eds.: D. Drabløs and A. Tollan. SNSF-project, Oslo—Ås.

— and O. K. SKOGHEIM. 1982. Physiological stress and mortality of Atlantic salmon, *Salmo salar* L. in acid water with high levels of aluminium. ICES, C.M 1982/M: 29. 15 p.

— and O. K. SKOGHEIM. 1984. A comparative study on salmonid fish species in acid aluminium-rich water. II. Physiological stress of one- and two-year-old fish. Rep. Inst. Freshw. Res., Drottningholm 61: 186—194.

ROSSELAND, L. 1953. Om virksomheten i 1948, p. 30. (Annual report 1948. Annual Report from the Fishery Inspector to the Department of Agriculture for the years 1948—50.) Oslo, Norway. (In Norwegian.)

SCHOFIELD, C. L. 1977. Acid snow-melt effects on water quality and fish survival in the Adirondack Mountains of New York State. Res. Proj. Tech. Comp. Rep. Project A-072-NY. Cornell Univ., Ithaca, New York. 27 p.

SEIP, H. M., L. MÜLLER and A. NAAS. 1984. Aluminium speciation: Comparison of two spectrophotometric analytical methods and observed concentrations in some acidic aquatic systems in southern Norway. Wat., Air Soil Poll. 23: 81—95.

SNEKVIK, E. 1975. Episoder med fiskedød i forbindelse med forsuring av vassdrag i 1969. (Episodic fish-kill in 1969 in relation to acidification.) Dir. Wildl. Freshw. Fish., Rep. Fish. Res. Div. 9 p. (In Norwegian.)

STAINTON, M. P. 1974. An automated method for determination of chloride and sulphate in freshwater using cation exchange and measurement of electric conductance. Limnol. Oceanogr. 19: 707—711.

STATE POLLUTION CONTROL AUTHORITY. 1982. Overvåking av langtransportert forurenset luft og nedbør. (Monitoring program for long range transported pollutants in air and precipitation, Annual Report 1981.) Statens forurensningstilsyn. Rapp. 64/82. 176 p. (In Norwegian.)

WESTALL, J. C., J. L. ZACHARY and F. N. M. MOREL. 1976. MINEQL, a computer program for the calculation of chemical equilibrium composition of aqueous systems. Dep. Civil. Eng. MIT, Tech. Note 18. 91 p.

WRIGHT, R. F. and O. K. SKOGHEIM. 1983. Aluminium speciation at the interface of an acid stream and a limed lake. Vatten 39: 301—304.

Reports from the Institute of Freshwater Research, Drottningholm

*1. *Gunnar Alm.* Statens undersöknings- och försöksanstalt för sötvattensfisket. Dess tillkomst, utrustning och verksamhet. English summary. 1933. Pris kr. 0: 75.
*2. *Gunnar Alm.* Vätterns röding, Fiskeribiologiska undersökningar. Deutsche Zusammenfassung. 1934. Pris kr. 0: 75.
*3. *Christian Hessle.* Märkningsförsök med gädda i Östergötlands skärgård åren 1928 och 1930. English summary. 1934. Pris kr. 0: 50.
4. *Gottfrid Arvidsson.* Märkning av laxöring i Vättern. Deutsche Zusammenfassung 1935. Pris kr. 0: 75.
*5. *Sten Vallin.* Cellulosafabrikerna och fisket. Experimentella undersökningar. Deutsche Zusammenfassung. 1935. Pris kr. 0: 75.
*6. *Gunnar Alm.* Plötsliga temperaturväxlingars inverkan på fiskar. Deutsche Zusammenfassung. 1935. Pris kr. 0: 75.
*7. *Christian Hessle.* Gotlands havslaxöring. English summary. 1935. Pris kr. 0: 75.
*8. *Orvar Nybelin.* Untersuchungen über den bei Fischen krankheitserregenden Spaltpilz Vibrio Anguillarum. 1935. Pris kr. 1: 25.
9. *Orvar Nybelin.* Untersuchungen über die Ursache der in Schweden gegenwärtig vorkommenden Krebspest. 1936. Pris kr. 0: 75.
10. *E. Rennerfelt.* Untersuchungen über die Entwicklung und Biologie des Krebspestpilzes Aphanomyces astaci. 1936. Pris kr. 0: 75.
11. *Gunnar Alm.* Huvudresultaten av fiskeribokföringsverksamheten. Deutshe Zusammenfassung. 1936. Pris kr. 1: —.
12. *Gunnar Alm.* Industriens fiskeavgifter och deras användning. 1936. Pris kr. 1: 50.
*13. *H. Bergström och Sten Vallin.* Vattenförorening genom avloppsvattnet från sulfatcellulosafabriker. 1937. Pris kr. 0: 75.
*14. *Gunnar Alm.* Laxynglets tillväxt i tråg och dammar. English summary. 1937. Pris kr. 0: 75.
*15. *Gunnar Alm.* Undersökningar över tillväxt m.m. hos olika laxöringformer. English summary. 1939. Pris kr. 2: 50.
16. *Lars Brundin.* Resultaten av under perioden 1917—1935 gjorda fiskinplanteringar i svenska sjöar. Deutsche Zusammenfassung. 1939. Pris kr. 1: —.
17. *Nilss Törnquist.* Märkning av vänerlax. English summary. 1940. Pris kr. 1: —.
18. *Sven Runnström.* Vänerlaxens ålder och tillväxt. English summary. 1940. Pris kr. 1: —.
19. *Arne Lindroth.* Undersökningar över befruktnings- och utvecklingsförhållanden hos lax (Salmo salar). Deutsche Zusammenfassung. 1942. Pris kr. 0: 75.
*20. *Lars Brundin.* Zur Limnologie jämtländischer Seen. 1942. Pris kr. 2: —.
*21. *Gunnar Svärdson.* Studien über den Zusammenhang zwischen Geschlechtsreife und Wachstum bei Lebistes. 1943. Pris kr. 1: —.
*22. *Gunnar Alm.* Befruktningsförsök med laxungar samt laxens biologi före utvandringen. (Fertilization-Experiments with Salmon-parr.) English summary. 1943. Pris kr. 1: 50.
*23. *Gunnar Svärdson.* Chromosome Studies on Salmonidae. 1945. Pris kr. 3: —.

24. *Arne Lindroth.* Zur Biologie der Befruchtung und Entwicklung beim Hecht. (Gäddans befruktnings- och utvecklingsbiologi samt gäddkläckning i glas.) 1946. Pris kr. 3: —.
25. *Gunnar Alm.* Reasons for the occurrence of stunted fish populations. (Uppkomsten av småväxta fiskbestånd, spec. hos abborre.) 1946. Pris kr. 3: —.
26. *Gösta Högström.* Olika impregneringsämnens lämplighet för grovgarnig fiskredskap. Deutsche Zusammenfassung. 1947. Pris kr. 1: —.
27. *A. Määr.* Über die Aalwanderung im Baltischen Meer auf Grund der Wanderaalmarkierungsversuche im finnischen und livischen Meerbusen i. d. J. 1937—1939. 1947. Pris kr. 2: —.
28. *Elias Dahr.* Biologiska studier över siken vid mellansvenska östersjökusten. English summary. 1947. Pris kr. 2: —.
29. Annual Report for the Year 1948 and Short Papers. 1949. Pris kr. 3: 50.
30. *Lars Brundin.* Chironomiden und andere Bodentiere der südschwedischen Urgebirgsseen. English summary. 1949. Pris kr. 15: —.
*31. Annual Report for the Year 1949 and Short papers. 1950. Pris kr. 3: 50.
32. Annual Report for the Year 1950 and Short papers. 1951. Pris kr. 4: 50.
33. Annual Report for the Year 1951 and Short Papers. 1952. Pris kr. 8: —.
34. Annual Report for the Year 1951 and Short Papers. 1953. Pris kr. 8: —.
35. Annual Report for the Year 1951 and Short Papers. 1954. Pris kr. 8: —.
36. Annual Report for the Year 1951 and Short Papers. 1955. Pris kr. 8: —.
*37. Report from the Institute of Freshwater Research, Drottningholm. 1956. Pris kr. 8: —.
*38. Report from the Institute of Freshwater Research, Drottningholm. 1957. Pris kr. 8: —.
39. Report from the Institute of Freshwater Research, Drottningholm. 1958. Pris kr. 8: —.
*40. Report from the Institute of Freshwater Research, Drottningholm. 1959. Pris kr. 8: —.
41. Report from the Institute of Freshwater Research, Drottningholm. 1960. Pris kr. 8: —.
42. Report from the Institute of Freshwater Research, Drottningholm. 1961. Pris kr. 8: —.
*43. *Lars B. Höglund.* The Reactions of Fish in Concentration Gradients. 1961. Pris kr. 8: —.
44. Report from the Institute of Freshwater Research, Drottningholm. 1962. Pris kr. 8: —.
45. Report from the Institute of Freshwater Research, Drottningholm. 1964. Pris kr. 8: —.
*46. Report from the Institute of Freshwater Research, Drottningholm. 1965. Pris kr. 12: —.
*47. Report from the Institute of Freshwater Research, Drottningholm. 1967. Pris kr. 25: —.
*48. Report from the Institute of Freshwater Research, Drottningholm. 1968. Pris kr. 25: —.
49. Report from the Institute of Freshwater Research, Drottningholm. 1969. Pris kr. 25: —.
50. Report from the Institute of Freshwater Research, Drottningholm. 1970. Pris kr. 30: —.
51. Report from the Institute of Freshwater Research, Drottningholm. 1971. Pris kr. 30: —.
52. Report from the Institute of Freshwater Research, Drottningholm. 1972. Pris kr. 40: —.
53. Report from the Institute of Freshwater Research, Drottningholm. 1973. Pris kr. 40: —.
54. Report from the Institute of Freshwater Research, Drottningholm. 1975. Pris kr. 40: —.
55. Report from the Institute of Freshwater Research, Drottningholm. 1976. Pris kr. 40: —.
56. Report from the Institute of Freshwater Research, Drottningholm. 1977. Pris kr. 50: —.
57. Report from the Institute of Freshwater Research, Drottningholm. 1979. Pris kr. 50: —.
58. Report from the Institute of Freshwater Research, Drottningholm. 1979. Pris kr. 60: —.
59. Report from the Institute of Freshwater Research, Drottningholm. 1981. Pris kr. 60: —.
60. Report from the Institute of Freshwater Research, Drottningholm. 1982. Pris kr. 80: —.
61. Report from the Institute of Freshwater Research, Drottningholm. 1984. Pris kr. 80: —.

---

* Out of print.