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by

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## INTRODUCTORY COMMENTS

There is clear evidence that Atlantic salmon rivers in parts of the Maritimes have been and are being affected by increased acidification of precipitation. The recent stalemate in bilateral negotiations between Canada and the U.S., however, emphasizes the necessity for better documentation of the link between acidic precipitation and its deleterious effects on salmon populations.

The Westfield River project, as originally conceived, was designed to answer three questions: 1) How are migratory fishes, particularly Atlantic salmon, affected by low pH at various stages of their life cycles? 2) How is fish production in these streams altered when these species disappear? and 3) What is the source of acidity that is responsible for the decrease in pH in these rivers? The Westfield River was chosen because its pH regime (annual mean ca. 5.1) was thought to be near the minimum tolerable for salmon reproduction. There were some electrofishing data obtained 15 yr ago, indicating that it was one of the more productive survey areas in the Medway system at that time; and it was of suitable size for monitoring fish movements.

Fisheries research on the Westfield River was begun in the fall of 1980, preceding implementation of the Departmental acid rain program, when salmon eggs were planted in artificial redds in the Westfield (and several other streams) to determine the influence of the chemistry of redd interstitial water on survival of eggs and fry.

Due to the inevitable delays in hiring personnel, construction of field equipment, procuring lodging, etc., the full-scale field program was not begun until August of 1981 with the installation of counting fences, discharge gauging stations, and the initiation of fish, invertebrate and periphyton sampling programs. The field program has thus been in full operation for 20 mo with only one complete field season of data collection (1982) accomplished. Permanent field equipment now includes wet precipitation sampling equipment at three sites in the Westfield drainage basin, discharge gauging stations on the Westfield and its two main tributaries, and two counting fences (operational from April-December) situated at the two boundaries of the ½-km stretch of study area, and a stream-side trailer supplied with electricity and a source of Westfield River water for stream-side experimental set-ups. Precipitation and stream water chemistry of the Westfield and the two tributaries are monitored on a weekly basis with more frequent sampling during the study of specific precipitation episodes. The past winter was mild enough that monthly assessment of fish populations could be made by electrofishing.

In addition to the field program, a number of laboratory projects were initiated to provide some backup information to the field program. These projects included studies on the influence of low pH on the smoltification process, on egg, alevin and fry development, on sexual maturation and steroid production of Atlantic salmon adults, and on territorial and feeding behavior of salmon parr.

Findings of the field studies to date are:  
1) Survival of salmon eggs and fry in the Westfield system is inversely correlated with pH of the redd

interstitial water. 2) Stream-side experiments have demonstrated very high (ca. 70%) mortality of newly feeding fry in Westfield River water, and that this mortality could be eliminated by raising the river water to a pH of 6.0 by passing it through limestone. 3) Possibly as a result of this increased mortality, densities of yearling salmon in the Westfield are extremely low (migration of fry from the study areas is an alternate possibility). 4) Mature adult salmon returned to the Westfield River in 1982 in very low numbers, with most of the run (70%) consisting of strays from the salmon enhancement program initiated further upstream on the main Medway. 5) Numbers and biomasses of other migratory species (alewife and eel) utilizing, or migrating through the study area have also been estimated for one year, the eel population comprising most of the fish biomass in the study area.

The laboratory studies have demonstrated that 1) pH levels within ranges encountered in the Westfield drainage can decrease ion uptake and water uptake during hardening in Atlantic salmon alevins and fry. 2) Steroid production or release is impaired in male salmon during residence in the Westfield, possibly resulting in decreased egg fertilization. 3) Smoltification is impaired resulting in reduced ionic regulation and salinity tolerance. 4) Hatching can be delayed at low pH due to inhibition of the hatching enzyme and changes in egg capsule structure.

Several of the past studies (the smoltification, early developmental and stream behavior studies) are in the final stages of investigation and are scheduled to terminate in 1983-84.

We feel that it is important to continue the studies initiated on the fish populations of the Westfield River as less than 2 yr of research effort have been expended to date, while 4 yr are required just to monitor the salmon population through one generation. The Westfield acid rain study is also of strategic importance as the findings in this study will complement those of the nearby Kejimikujik Calibrated Watershed Study. The Westfield is a tributary of the Medway River and the findings will also be of use to management of the Medway salmon enhancement program as it provides information on movements of salmon studied in this program which return to the Westfield, and on the likelihood of enhancement success in four tributaries of the Medway.

The ensuing proceedings of the second Westfield Workshop detail the findings of the various studies for 1982.

R. H. Cook  
Fisheries and Environmental Sciences  
April 15, 1983.

## ABSTRACT

Peterson, R. H., and H. H. V. Hord [Eds.]. 1983. 1983 Workshop on acid rain. Can. Tech. Rep. Fish. Aquat. Sci. 1213: v + 79 p.

This report is a record of the papers presented at a Workshop organized by the Director of the St. Andrews Biological Station to review the results of research on acid rain carried out during 1982 under the aegis of the Fisheries and Environmental Sciences Division of the Fisheries Research Branch, Scotia-Fundy Region, Department of Fisheries and Oceans.

## RÉSUMÉ

Peterson, R. H., and H. H. V. Hord [Eds.]. 1983. 1983 Workshop on acid rain. Can. Tech. Rep. Fish. Aquat. Sci. 1213: v + 79 p.

Ce rapport est un dossier des communications présentées à un atelier organisé par le directeur de la station de biologie de St. Andrews pour faire le compte rendu des résultats de recherches sur les pluies acides conduites durant 1982 sous l'égide de la division des pêches et sciences de l'environnement de l'office des recherches sur les pêcheries, région Scotia-Fundy, ministère des pêches et océans.



WATER CHEMISTRY AND ECOLOGY OF FISH POPULATIONS  
IN STREAMS OF THE WESTFIELD DRAINAGE IN NOVA SCOTIA

by

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## ABSTRACT

The chemical composition of precipitation and of water from study streams in the Westfield River drainage and the ecological studies of fish populations conducted in these streams during 1982 are summarized. The Westfield River had a mean annual pH of 4.8; tributary streams had mean annual pH's <4.6. Mature adult salmon returned to the Westfield River in very low numbers, with most of the run (ca. 70%) consisting of strays from the salmon enhancement program initiated on the Medway River. Possibly as a result of increased mortality, densities of yearling salmon in the Westfield River were extremely low; no juvenile salmon were found in the more acid tributary streams. Observed differences in growth of juvenile salmon in streams of different pH could be explained as density-dependent effects. Successful emergence of salmon fry from natural redds was confirmed. However, streamside experiments demonstrated very high (ca. 70%) mortality of newly feeding salmon fry in the Westfield River water and this mortality could be eliminated by raising the river water to pH 6.0 by passing it through limestone.

## RÉSUMÉ

Ce qui suit est un résumé de la composition chimique des précipitations et de l'eau de cours d'eau du bassin hydrographique de la rivière Westfield, ainsi que des études écologiques des populations de poissons menées dans ces cours d'eau en 1982. Le pH annuel moyen de la rivière Westfield est de 4,8; ceux des tributaires sont inférieurs à 4,6. Les saumons adultes matures ne retournent à la rivière Westfield qu'en très petit nombre, la plupart des remontes (ca 70%) étant des vagabonds en provenance du programme de revalorisation du saumon entrepris sur la rivière Medway. La densité des saumons d'un an dans la rivière Westfield est extrêmement faible, possiblement à cause de fortes mortalités; aucune saumoneau n'a été trouvé dans les tributaires, plus acides. Les différences de croissance observées chez les jeunes saumons dans des cours d'eau de pH différents pourraient peut-être s'expliquer en termes de dépendance de la densité. On a pu confirmer le succès de l'émergence des alevins de saumon des frayères naturelles. Cependant, des expériences menées sur le terrain ont démontré de très fortes (ca 70%) mortalités d'alevins qui commençaient à se nourrir dans l'eau de la rivière Westfield. Ces mortalités pourraient être éliminées en élevant le pH de l'eau de la rivière à 6,0, en la faisant passer à travers du calcaire.

## A. PRECIPITATION AND STREAM WATER CHEMISTRY

Between November 1981, and October 1982, the total precipitation (rain and snow) in the Westfield drainage was 1449 mm. The wettest months were November, December, April and June. For the same period, bulk precipitation in the Westfield drainage had an annual mean pH of 4.78.  $\text{Na}^+$  was the dominant cation and  $\text{Cl}^-$  was the dominant anion present, an indication of the marine influence on precipitation (Table 1). Other dominant ions were:  $\text{H}^+$  for cations and  $\text{SO}_4^{2-}$  for anions. The pH of bulk precipitation fluctuated throughout the year but was lowest in November and March (Fig. 1).

In the Westfield River, the major decrease in pH recorded in the fall and winter of 1981 did not occur in 1982. The total monthly precipitation was always less than 100 mm during the fall of the latter. Instead, the river pH gradually increased from a low of 4.6 in March to a maximum of 5.3 in October (Fig. 1). The three streams monitored in the Westfield drainage were dominated by  $\text{Na}^+$  and  $\text{Cl}^-$  ions (Table 2). Both  $\text{H}^+$  and  $\text{SO}_4^{2-}$  were highest in the headwater streams (Round Lake Brook and Moose Pit Brook) of the Westfield River, and the annual mean pH was less than 4.6 in both brooks. The average pH of the Westfield River was approximately equal to that of precipitation. The mean concentrations of  $\text{Ca}^{++}$  and  $\text{Mg}^{++}$  were relatively higher in all three streams than in the atmospheric inputs.  $\text{NH}_4^+$ ,  $\text{NO}_3^-$  and  $\text{PO}_4^{3-}$  concentrations were extremely low in the three streams, usually less than  $0.01 \text{ mg L}^{-1}$ .

The highest Al concentrations were found in the low pH headwater streams (Fig. 2). Fe concentration was highest in Moose Pit Brook, a low order stream with a high organic content and a drainage comprised of bogs. Both other streams have headwater lakes which are likely to have a dampening effect on outputs. Temporal variations in Al and Fe concentrations are apparently related to streamflow; concentrations were usually highest during periods of low flow such as from July to October.

Al in all three streams likely occurs as non-labile, organically complexed Al since concentrations of humates were high:  $9 \text{ mg L}^{-1}$  in the Westfield River,  $11 \text{ mg L}^{-1}$  in Round Lake Brook, and  $20 \text{ mg L}^{-1}$  in Moose Pit Brook.

## B. ECOLOGY OF FISH POPULATIONS

### 1) Estimated numbers of migrating fishes

Weirs for counting migrating fishes were operated at the boundaries of the Westfield River study site from April to December during 1982. Water flow was low ( $\leq 10 \text{ m}^3 \text{ s}^{-1}$ ) throughout this period.

Extremely low water level from mid-July to November effectively prevented Atlantic salmon from migrating to the headwaters of the Westfield River to spawn. A total of five salmon (all of hatchery origin) and 34 grilse (23 of which were of hatchery origin) migrated to and past the study area; all but three of these fish were counted in late June-early July. However, some salmon succeeded in migrating to the lower stretches of the Westfield River to spawn during November; at least 30 redds were completed there by salmon.

Low water in the Westfield River, however, did not preclude alewives from migrating to the headwaters. A total of 4,434 alewives were counted migrating upstream to spawn between May 10 and July 10. Most of these were ripe (93%), and 48% were females. Spent alewives were captured returning downstream from June 11 to September 12; 2,354 fish were counted (53% survival of spawners.). The emigration of juvenile alewives from the system was continuous from July 30 to November 26. During this period a barrier net was in place over the weir to maximize the capture rate of these small fish; without the barrier net the weir was only 40% efficient for capturing juvenile alewives. A total biomass output of 1,050 kg (wet weight) was estimated for juvenile alewives ( $N = 177,077$  fish).

Few salmon smolts ( $N = 46$ ) were captured moving downstream during the spring (April 26-May 29). There was also no appreciable migration of parr (yearlings or older) at that time or during the fall. The weirs proved to be unreliable for monitoring the dispersal or the movements of under-yearlings during their first season. Emigration of 0+ parr to downstream areas (i.e. Medway River) could not be ruled out as a possible explanation for the extremely low older parr densities in the Westfield River and apparent lack of smolts.

### 2) Estimated densities and growth of juvenile Atlantic salmon

Six streams in mainland Nova Scotia were electrofished in 1982. In three of the rivers surveyed, no juvenile salmon were found. The streams, Round Lake Brook, Halfway Brook, and Moose Pit Brook, are tributaries to the Westfield River and each is consistently below pH 4.9. American eel is the most abundant species in these brooks; they also support a few brook trout, yellow perch, white sucker, ninespine stickleback, and banded killifish.

The other sites were on the Westfield River, the North River, and Harmony Brook. The North River, a tributary to the LaHave River, is highly colored with  $\text{SO}_4^{2-}$  concentration similar to the Westfield; however,  $\text{Ca}^{++}$  concentration is double that in the Westfield and a pH near 6.0 is usual. Harmony Brook, a tributary to the Medway River, is also usually above pH 6.0; however, unlike the Westfield and North Rivers, it is a clear water stream with almost half of the  $\text{SO}_4^{2-}$  concentration of the other rivers.

Density estimates of each species were made regularly at sites on the Westfield and North Rivers by the catch-per-unit-effort method. Growth parameters were also measured. Juvenile salmon have only recently (1981-1982) been introduced by limited stocking in Harmony Brook, a natural barrier at the mouth of the brook used to prevent access by Atlantic salmon. Density estimates of juvenile salmon are not yet possible in this brook because of the very low numbers, but growth parameters were measured.

Preliminary density estimates are presented only for juvenile salmon (Fig. 3). Extrapolation of the survivorship curve between the 0+ and 1+ age-classes is but an assumption since estimates are only available for a single year. Densities of 0+ salmon in the Westfield River were lower than in the North River throughout the first season even though

the intensity of spawning the previous fall was high in both areas. Only a few older juveniles remain in the Westfield River and a high overwinter mortality (or emigration?) is assumed in comparison to that in the North River. The increase in density of 1+ parr in the North River during November results from immigration of precocious males to the area. The average biomass of 0+ parr in the Westfield is about 70% of that in the North River, whereas that of 1+ parr is reduced to less than 10% (Table 3).

The growth of 0+ parr in the North River was almost identical to that in the more acidic Westfield River (Fig. 4). The faster growth of this age-class in Harmony Brook could result from the low density of this age-class in the brook; only 2000 fry were planted in July 1982. Older parr (1+) were considerably larger, throughout the season, in the Westfield River and Harmony Brook than in the North River, and the rate of growth was faster. This difference can most likely be attributed to much lower densities of 1+ parr in the former two streams. The sudden peaks in the growth curves of older parr during the fall result from the immigration of larger fish (mostly precocious males) to the study sites at that time. Observed differences in the growth of juvenile salmon can, thus far, be explained as density-dependent effects; they do not seem to be related directly to the different water chemistry of the rivers compared.

### 3) Effects of Westfield River water on Atlantic salmon fry

An initial study in 1981 determined that hatchability of salmon eggs in the Westfield River was greater than 50%. Subsequently, successful emergence of salmon fry from natural redds was confirmed, using emergence traps. The high densities of fry observed in the vicinity of redds after swim-up in June 1982 suggests that survival to the initial period of exogenous feeding may be adequate.

The effects of Westfield River water on salmon fry were investigated in a streamside experiment during 1982. Salmon fry were reared from swim-up (May 23) for a 2-mo period in tanks (22 L) receiving unaltered water from the Westfield River and river water percolated through a limestone filter. There were 750 fry in each of four replicates (tanks) for each treatment, a total of 3000 fry/treatment. Flow was controlled at 2 L min<sup>-1</sup> in each tank and water depth maintained at 7.5 cm. The fry were fed regularly from the onset in amounts approximating 5% of body weight each day.

The river water was consistently near pH 5.0 while treatment with limestone maintained the water at pH 6.0 throughout the experiment. Mortality and growth data are summarized in Fig. 5, and preliminary data on water and body ion content are displayed in Fig. 6. Data for four replicates at each pH regime were combined since there were no significant differences ( $P = 0.05$ ).

Initially, fry in both treatments fed, although not as voraciously at pH 5 as at pH 6. There was nevertheless an initial weight loss at both pH's as fry adapted to exogenous feeding. After 10 d, fry at pH 6 started growing rapidly while at pH 5 there was a continued weight loss until high mortalities ensued. Mortality at pH 5 occurred mostly between day 15 and 25; both 50% cumulative mortality and

peak mortality were on day 21. The condition of fry was below 0.6 when mortality started and all dead fry weighed less than 200 mg.

The total cumulative mortality for the 54-d period was over 70% at pH 5 and less than 5% at pH 6. The relatively high aluminum concentrations (range 170-230  $\mu\text{g Al L}^{-1}$ ) were almost identical at both pH regimes.

Surviving fry after 25-30 d at pH 5 were growing very rapidly and the condition for these fish exceeded that of fry at pH 6. Eventually, after 40 d, fry at both pH levels had similar growth rates and condition factors.

The ionic composition of water and of fry (with stomach excised) in both treatments was compared regularly throughout the experiment. There were no differences in water nor in body  $\text{Mg}^{++}$  and  $\text{K}^{+}$  content between treatments and concentrations were relatively constant over the test period. Water  $\text{Mg}^{++}$  concentration was 35  $\mu\text{eq L}^{-1}$  and  $\text{K}^{+}$  was 5  $\mu\text{eq L}^{-1}$ , total body  $\text{Mg}^{++}$  range was 0.10-0.15  $\mu\text{eq mg dry wt}^{-1}$  and  $\text{K}^{+}$  was 0.25-0.30  $\mu\text{eq mg dry wt}^{-1}$ .

Water  $\text{Na}^{+}$  concentration did not differ between treatments, whereas  $\text{Ca}^{++}$  was more than doubled by filtering river water through limestone. At the onset, total body  $\text{Na}^{+}$  and  $\text{Ca}^{++}$  contents were similar at both pH's, and the concentration of both ions increased during the period of weight loss. The body  $\text{Na}^{+}$  and  $\text{Ca}^{++}$  contents of fry at pH 5 were both consistently lower than those of fry at pH 6 for approximately 10 d preceding the mortality period. Body  $\text{Na}^{+}$  content remained lower for fry at pH 5 until mortality ceased. Thereafter, surviving fry at pH 5 and fry at pH 6 had similar total body  $\text{Na}^{+}$  and  $\text{Ca}^{++}$  concentrations.

The untreated river water had a severe impact on salmon fry during the initial feeding period, 15-25 d after emergence. Such mortalities are likely to severely limit salmon recruitment in the Westfield River.

Table 1. Chemical data of wet precipitation, Westfield River drainage area, November 1981-October 1982. Annual mean (unweighted) is calculated from weekly bulk samples.

	$\mu\text{eqL}^{-1}$	% eq
Na	28.8	48.8
Ca	4.6	7.8
Mg	6.7	11.4
K	2.4	4.1
H	16.5	27.9
Cations	59.0	100.0
Cl	36.7	63.8
SO <sub>4</sub>	20.8	36.2
Anions	57.5	100.0
Total ions	116.5	
Anions/cations	0.97	
<hr/>		
$\text{pH} = -\log(\Sigma 10^{-\text{pH}}/N)$	4.78	
Alkalinity ( $\text{mgL}^{-1} \text{CaCO}_3$ )	-0.72	
Acidity ( $\mu\text{eqL}^{-1}$ )	89.3	
Total precipitation (mm)	1449	

Table 2. Chemical composition of study streams, Westfield River drainage area, November 1981-October 1982. Annual mean (unweighted) is calculated from weekly values for grab samples.

	Westfield River		Round Lake Brook		Moose Pit Brook	
	$\mu\text{eqL}^{-1}$	% eq	$\mu\text{eqL}^{-1}$	% eq	$\mu\text{eqL}^{-1}$	% eq
Na	100.3	54.1	104.9	51.3	116.1	50.6
Ca	31.4	16.9	31.9	15.6	40.4	17.6
Mg	32.4	17.5	32.4	15.8	41.3	18.0
K	6.6	3.6	8.9	4.3	5.8	2.5
H	14.8	7.9	26.6	13.0	26.0	11.3
Cations	185.5	100.0	204.7	100.0	229.6	100.0
Cl	110.3	58.9	116.9	57.3	150.6	61.3
SO <sub>4</sub>	77.1	41.1	87.2	42.7	95.2	38.7
Anions	187.4	100.0	204.1	100.0	245.8	100.0
Total ions	372.9		408.8		475.4	
Anions/cations	1.01		1.00		1.07	
<hr/>						
$\text{pH} = -\log(\Sigma 10^{-\text{pH}}/N)$	4.83		4.57		4.58	
Alkalinity ( $\text{mgL}^{-1} \text{CaCO}_3$ )	-0.47		-1.16		-0.90	
Acidity ( $\mu\text{eqL}^{-1}$ )	130.4		164.7		214.6	

Table 3. Average biomass (g wet weight  $m^{-2}$ ) of juvenile salmon during the growing season, 1982; biomass at the end of the growing season (November) in brackets.

Site	Age-class		Total
	0+	1+	
Westfield River	0.23 (0.23)	0.07 (0.03)	0.30 (0.26)
North River	0.32 (0.32)	0.83 (1.72)	1.15 (2.04)

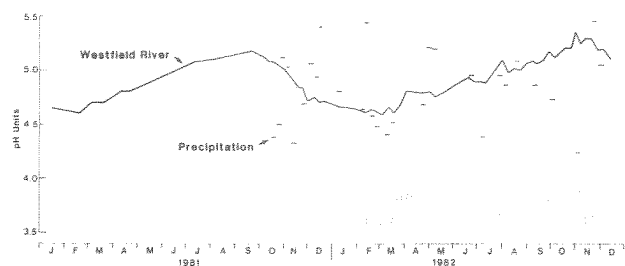


Fig. 1. Seasonal variation in pH of precipitation and of the Westfield River during 1981-1982.

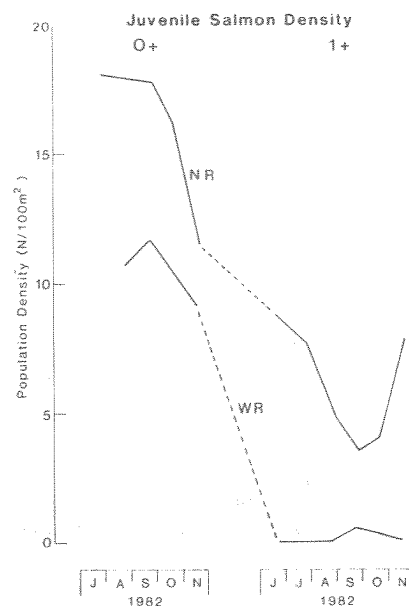


Fig. 3. Juvenile Atlantic salmon densities in the Westfield River (WR) and in the North River (NR) during 1982.

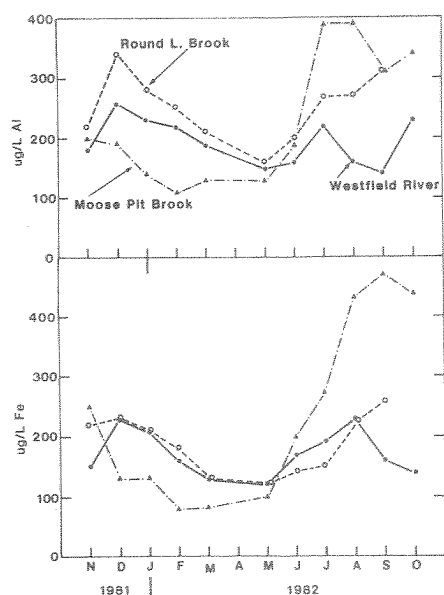


Fig. 2. Seasonal variation in Al and Fe concentrations of the three study streams from November, 1981, to October, 1982.

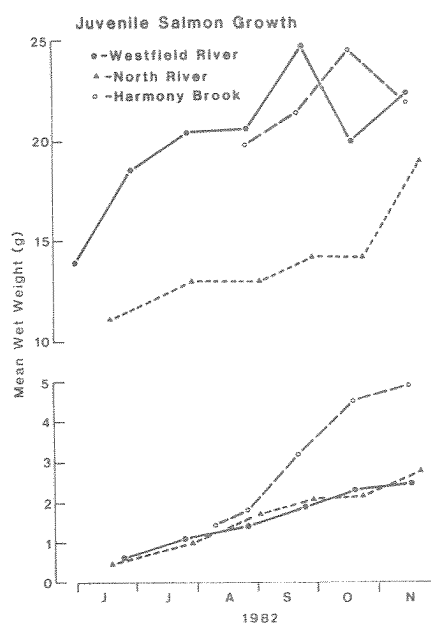


Fig. 4. Increase in mean weight of 0+ (lower curves) and 1+ (upper curves) Atlantic salmon parr from the three study streams during 1982.

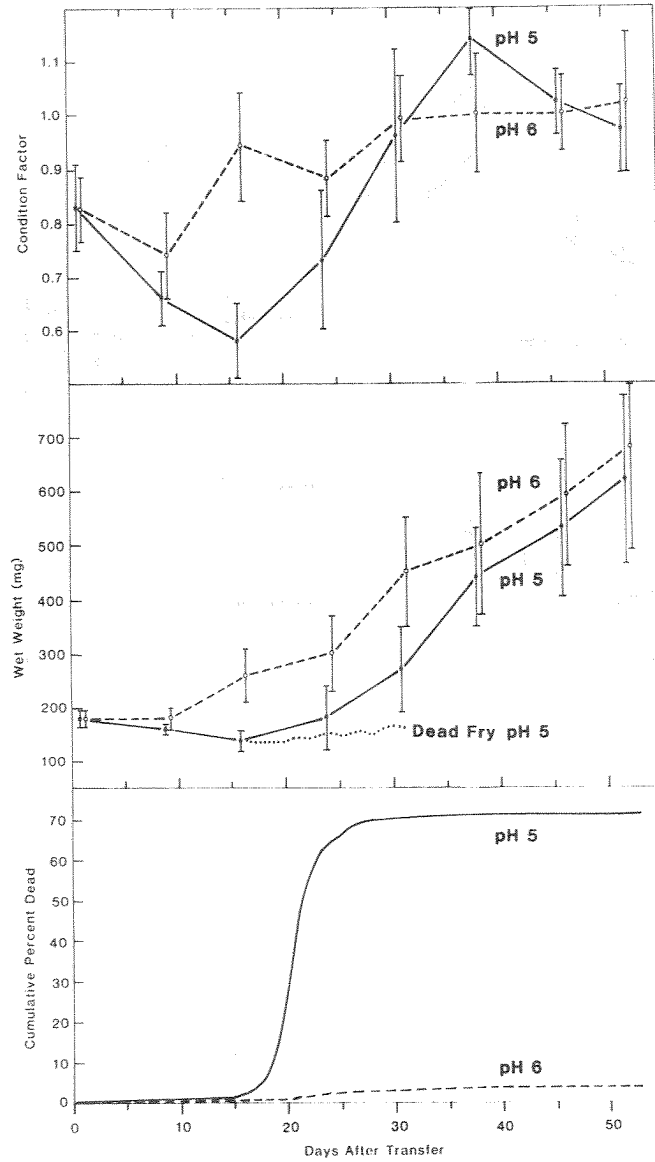


Fig. 5. Mortality, growth, and condition of Atlantic salmon fry reared at pH 5.0 and 6.0 from swim-up (day 0); means  $\pm$  1 standard deviation are shown for weight and condition.

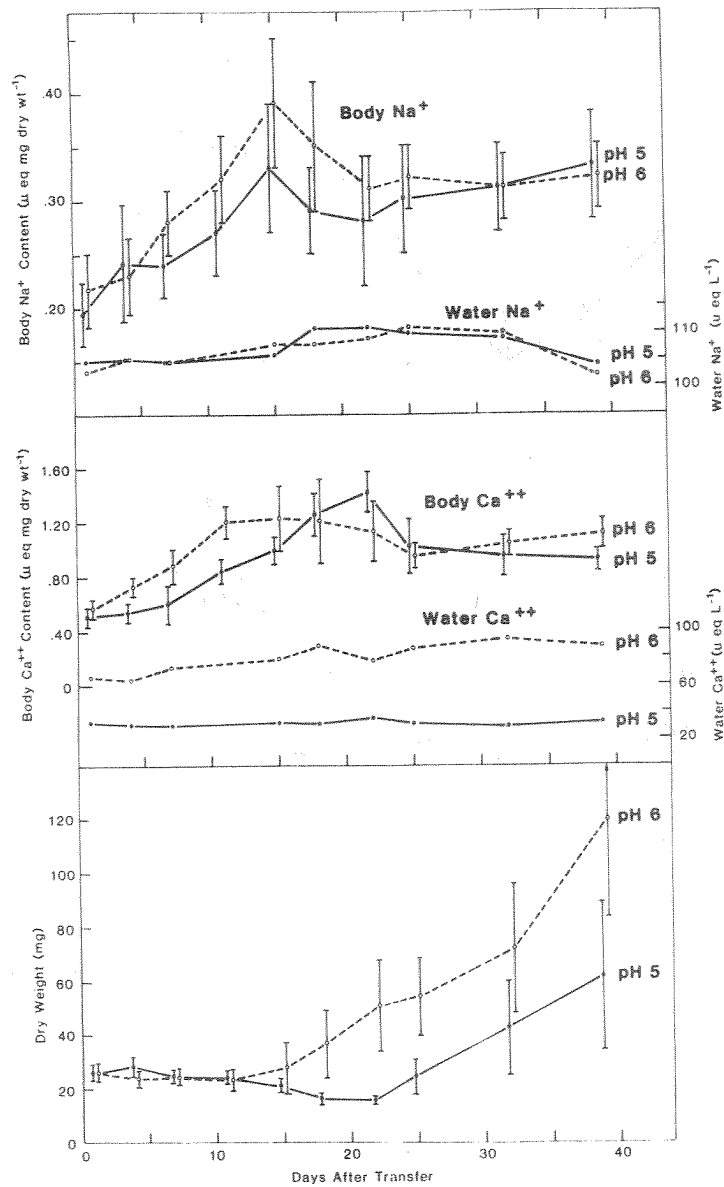


Fig. 6. Dry weight, and body  $\text{Na}^+$  and  $\text{Ca}^{++}$  content of Atlantic salmon fry reared at pH 5.0 and 6.0 from swim-up (day 0); means + 1 standard deviation are shown. Water  $\text{Na}^+$  and  $\text{Ca}^{++}$  concentrations at both pH's are also compared.



INVESTIGATION OF MACROINVERTEBRATES FROM THE WESTFIELD AND  
NORTH RIVER STUDY AREA, NOVA SCOTIA.

by

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## ABSTRACT

This is a preliminary report on a study of the benthic fauna of the Westfield and North Rivers containing information from collections made between April and December 1982. Representatives of the orders Diptera, Trichoptera, Ephemeroptera and Plecoptera (86-97%), dominated the benthic invertebrate population. Of these, 41-70% belonged to the family Chironomidae.

## RÉSUMÉ

Nous présentons ici un rapport préliminaire d'une étude de la faune benthique des rivières Westfield et North, incorporant des données recueillies entre avril et décembre 1982. La population d'invertébrés benthiques est dominée par des représentants des ordres Diptera, Trichoptera, Ephemeroptera et Plecoptera (86-97%). Parmi ceux-ci, 41-70% appartiennent à la famille des Chironomidae.

## INTRODUCTION

A complete study of an aquatic ecosystem should include an investigation of the benthic invertebrate fauna which serve as the main food source for many fish (Johnston 1980; Logan 1963).

The invertebrate benthic fauna of rivers have been examined to determine the taxonomic composition, diversity, relative abundance and seasonal variation (Babcock 1949; Minshall 1968; Minshall and Kuehne 1969; Egglisshaw 1980; Turcotte and Harper 1982).

The purpose of this preliminary report is to examine the benthic fauna of the Westfield and North Rivers, N.S., to determine their taxonomic composition, and study their seasonal variation from April-December 1982 in the Westfield River and June-December 1982 in the North River. This study is to be completed in June 1983 at which time the annual production for the two rivers will be estimated.

## METHODS

Invertebrates were collected from the Westfield and North Rivers, N.S., using a Surber sampler (Macan 1958), 30 cm by 30 cm with a mesh size of 1 mm, Coleman-Hynes pot samplers (Coleman and Hynes 1970), having a diameter of 10 cm and a depth of 20 cm, and drift nets with a 25 cm by 2.5 cm opening and a mesh size of 604  $\mu$ m.

Three transects were sampled in the Westfield River consisting of 9 pot sample sites and 9 Surber sample sites. One of the transects was selected as a production study site. Each pot sample site contained a duplicate set of pots and each Surber sample site consisted of 5 random samples. Three sites for pots and Surber samples in the North River were chosen over a distance of approximately 100 m.

Drift nets were set upstream and downstream of the production site in the Westfield River and only upstream of the production site in the North River. Sampling methods were the same as those described by Knox and Dadswell (1982). Samples were preserved (in 80% alcohol, 20% formalin solution), sorted, enumerated and identified to the lowest possible taxonomic group.

## RESULTS AND DISCUSSION

The orders Diptera, Trichoptera, Ephemeroptera and Plecoptera made up the largest percentage (86-97%), of invertebrates found in both pot and Surber samples from the Westfield and North Rivers (Table 1).

The Chironomidae were by far the most dominant invertebrate found in either river (41-70%), and Trichoptera were next in abundance ranging from 6-36%, while Ephemeroptera (5-9%) were slightly more abundant than Plecoptera (3-4%) (Table 1). The remaining invertebrates comprise only 4-13% of the total fauna collected (Table 1).

The total number and mean number per  $m^2$  of major groups of invertebrates collected with Surber sampler from the Westfield and North Rivers are

recorded in Table 2. The total number and mean number per pot sample for both rivers are recorded in Table 3.

Seasonal variation of the different orders collected is shown in Fig. 1 (Westfield River, Surber sample), Fig. 2 (North River, Surber sample), Fig. 3 (Westfield River, pot sample) and Fig. 4 (North River, pot sample). Since the data given are only for half a sampling year, these figures do not report the entire seasonal variation of the two rivers.

In order to estimate the annual production, using the Hynes Method (Hamilton 1969; Waters and Crawford 1973) the invertebrates were grouped into orders and then separated into 1-mm length categories. Wet weight, dry weight and ash-free dry weight (AFDW) were determined. To determine the AFDW, Benke and Wallace's (1980) method was followed. The annual production will be calculated when the study is completed in June 1983 (Appendix 1).

To date 250 (0<sup>+</sup> and 1<sup>+</sup>) salmon stomachs, collected from the Westfield and North Rivers and Harmony Brook, N.S., have been analyzed for food content. These findings plus those of stomachs to be analyzed at a later date will be reported in September 1983.

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Table 1. Total and percent composition of macroinvertebrate orders collected with Surber sampler from the Westfield and North Rivers, N. S. (June-December 1982).

	Total collected Westfield River	% of Total	Total collected North River	% of Total
Plecoptera	1 070	4	1 037	4
Ephemeroptera	1 692	7	2 377	9
Trichoptera	9 194	36	3 913	16
Coleoptera	324	1	1 138	4
Odonata	230	1	628	2
Megaloptera	215	1	291	1
Chironomid	10 600	41	13 139	52
Other Diptera	633	2	1 187	5
Miscellaneous	191	1	391	2
Oligochaeta	1 489	6	1 048	4

Total and percent composition of Macroinvertebrate Orders collected with pot samples from the Westfield River (April-December 1982) and North River (July-December 1982).

	Total collected Westfield River	% of Total	Total collected North River	% of Total
Plecoptera	1 053	4	427	3
Ephemeroptera	1 426	5	1 394	8
Trichoptera	3 261	12	904	6
Coleoptera	50	<1	77	<1
Odonata	63	<1	93	<1
Megaloptera	28	<1	24	<1
Chironomid	13 164	47	9 879	70
Other Diptera	8 041	29	294	2
Miscellaneous	305	1	498	4
Oligochaeta	743	3	492	4

Table 2. Composition of benthos collected with Surber sampler from the Westfield and North Rivers, N. S. (June-December 1982).

	Total number collected Westfield R.	Mean Nq. per M <sup>2</sup>	S. D.	Total number collected North R.	Mean Nq. per M <sup>2</sup>	S. D.
Leuctra sp.	589	70.5	115.6	449	53.7	26.5
Paracapnia sp.	376	45.0	66.9	312	37.3	45.2
Other Plecoptera	105	12.6	8.4	276	33.0	19.2
Ephemerella sp.	751	89.9	114.3	851	101.8	143.6
Stenonema sp.	670	80.2	79.0	405	48.5	41.3
Leptophlebiidae	267	32.0	39.1	865	103.9	127.6
Other Ephemeroptera	4	.5	.9	256	30.6	34.7
Cheumatopsyche sp.	2 097	250.9	182.7	534	63.9	84.2
Hydropsyche sp.	366	43.8	33.8	566	68.3	94.0
Brachycentrus sp.	5 736	686.4	500.8	266	31.8	20.0
Other Trichoptera	995	119.1	58.1	2 547	304.8	311.5
Anisoptera	171	20.5	14.4	387	46.3	34.8
Zygoptera	59	7.1	5.9	241	29.9	33.6
Megaloptera	215	25.7	8.0	291	34.8	26.4
Coleoptera	324	38.7	17.6	1 138	136.2	72.7
Chironomid (larvae)	10 600	1 268.5	992.3	13 139	1 572.3	972.0
Simuliidae (larvae)	20	2.4	3.0	6	.7	1.1
Oligochaeta	1 489	178.2	70.9	1 048	125.4	94.0
Miscellaneous	191	22.9	15.1	391	46.7	31.7
Other Diptera	613	73.4	62.8	1 181	141.3	119.8
Total	25 638	3 068.0	1 421.1	25 149	3 009.5	1 877.7

Table 3. Composition of benthos collected with pot sampler from Westfield (April-December 1982) and North Rivers (July-December 1982).

	Total number collected Westfield R.	Mean No. pot sample <sup>a</sup>	S. D.	Total number collected North R.	Mean No. pot sample <sup>a</sup>	S. D.
<i>Leuctra</i> sp.	626	14.9	30.2	118	3.9	3.4
<i>Paracapnia</i> sp.	226	5.4	5.9	183	6.1	8.7
Other Plecoptera	201	4.8	3.2	126	4.2	1.0
<i>Ephemerella</i> sp.	195	4.6	4.2	138	4.6	3.5
<i>Stenonema</i> sp.	377	9.0	6.1	424	14.1	7.8
Leptophlebiidae	847	20.2	20.4	649	21.6	2.7
Other Ephemeroptera	7	.2	.2	183	6.1	4.4
<i>Cheumatopsyche</i> sp.	842	20.0	23.9	73	2.4	2.5
<i>Hydropsyche</i> sp.	196	4.7	6.3	41	1.4	.9
<i>Brachycentrus</i> sp.	1 279	30.4	61.0	27	.9	.8
Other Trichoptera	944	22.5	25.2	763	25.4	10.8
Anisoptera	30	.7	.8	48	1.6	1.4
Zygoptera	33	.8	.8	45	1.5	1.3
Megaloptera	28	.7	.7	24	.8	1.0
Coleoptera	50	1.2	1.7	77	2.6	1.8
Chironomid (larvae)	13 164	313.4	237.5	9 879	329.3	204.9
Simuliidae (larvae)	7 112	169.3	441.4	3	.1	.1
Oligochaeta	743	17.7	15.1	498	16.6	12.3
Miscellaneous	305	7.3	6.1	492	16.4	21.2
Other Diptera	929	22.1	23.2	291	9.7	7.2
Total	28 134	669.9	459.9	14 082	469.4	211.9

<sup>a</sup>Volume of pot sample = .0016 m<sup>3</sup>

Appendix 1. Weights in mg of Ephemeroptera, Plecoptera and Trichoptera collected from the Westfield River (June 1982-December 1982).

Length (mm)	N	Wet weight	Dry weight	Ash weight	AFDW	Mean AFDW
EPHEMEROPTERA						
0-1	30	.53	-	-	-	-
1-2	472	27.81	6.88	.99	5.89	.012
2-3	309	92.30	15.30	1.30	14.00	.045
3-4	255	180.27	30.27	2.89	27.38	.107
4-5	136	243.04	34.71	4.22	30.49	.224
5-6	45	200.27	28.32	3.19	25.13	.558
6-7	25	120.12	20.65	1.67	18.98	.759
7-8	23	191.45	30.26	2.61	27.65	1.202
8-9	8	137.20	22.11	1.62	20.49	2.561
9-10	7	140.21	22.09	1.61	20.48	2.926
PLECOPTERA						
0-1	71	.68	.43	-	-	-
1-2	71	4.03	1.26	.11	1.15	.016
2-3	78	16.89	4.96	.57	4.39	.056
3-4	83	47.63	8.24	.78	7.46	.090
4-5	117	99.18	16.71	1.83	14.88	.127
5-6	174	207.03	41.19	3.58	37.61	.216
6-7	148	261.48	50.25	4.09	46.16	.312
7-8	54	168.25	32.93	2.35	30.58	.566
8-9	16	122.17	22.52	1.15	21.37	1.902
9-10	5	109.16	13.77	.49	13.28	2.656
TRICHOPTERA						
0-1	63	1.44	.10	-	-	-
1-2	350	23.41	5.77	.69	5.08	.015
2-3	1466	339.50	75.37	6.65	68.73	.047
3-4	422	323.87	48.71	1.69	47.02	.106
4-5	342	564.67	93.10	7.53	85.57	.250
5-6	140	344.94	58.86	4.99	53.87	.385
6-7	103	360.52	56.30	5.06	51.24	.497
7-8	59	285.16	65.72	5.09	60.63	1.028
8-9	45	235.59	39.77	3.30	36.47	.810
9-10	26	253.07	59.75	4.00	55.75	2.144



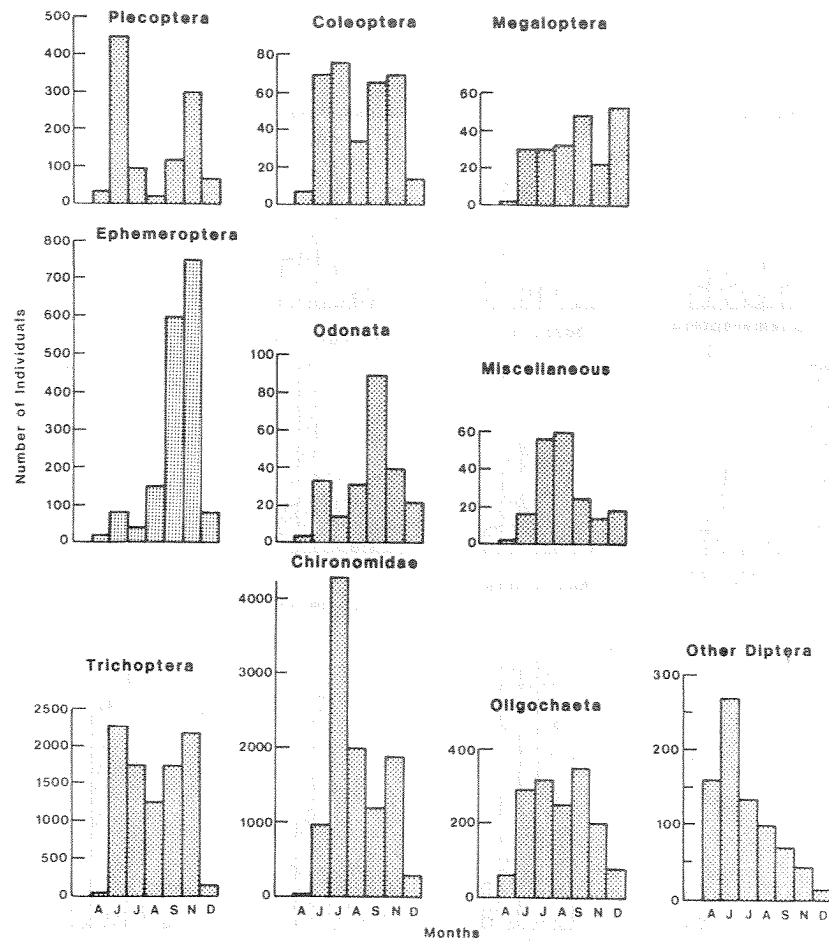


Fig. 1. Monthly variations in the density of the main groups of invertebrates collected from the Westfield River (numbers per 2.15 m<sup>2</sup>) 1982.

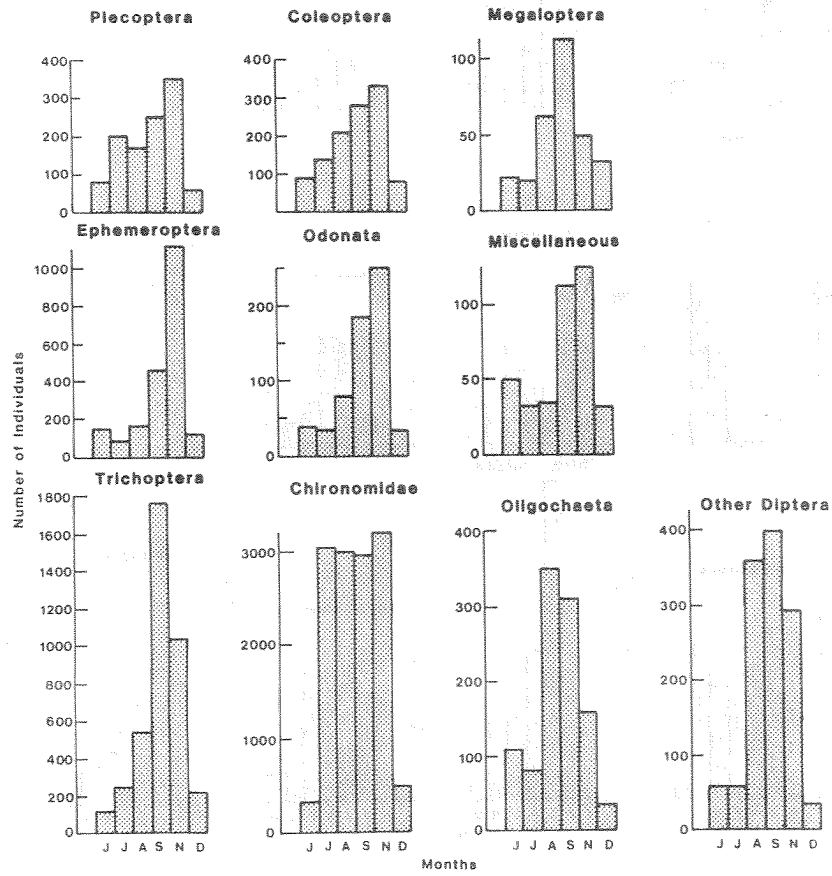


Fig. 2. Monthly variations in the density of the main groups of invertebrates collected from the North River (numbers per 2.15 m<sup>2</sup>) 1982.

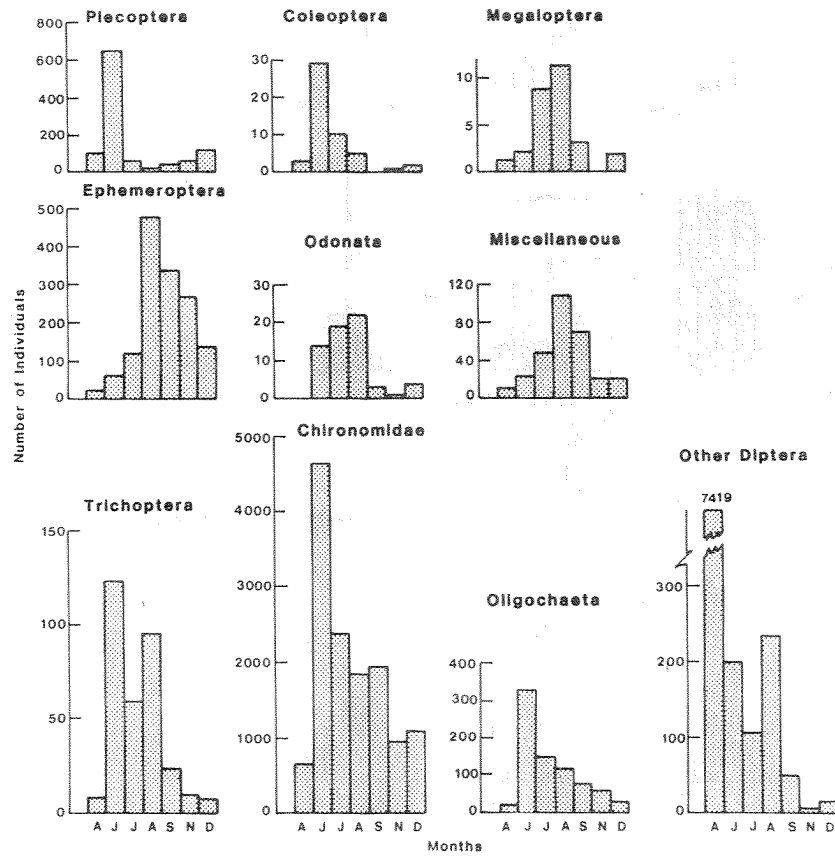


Fig. 3. Monthly variations in the density of the main groups of invertebrates collected from the Westfield River (numbers per 0.05 m<sup>2</sup>) 1982.

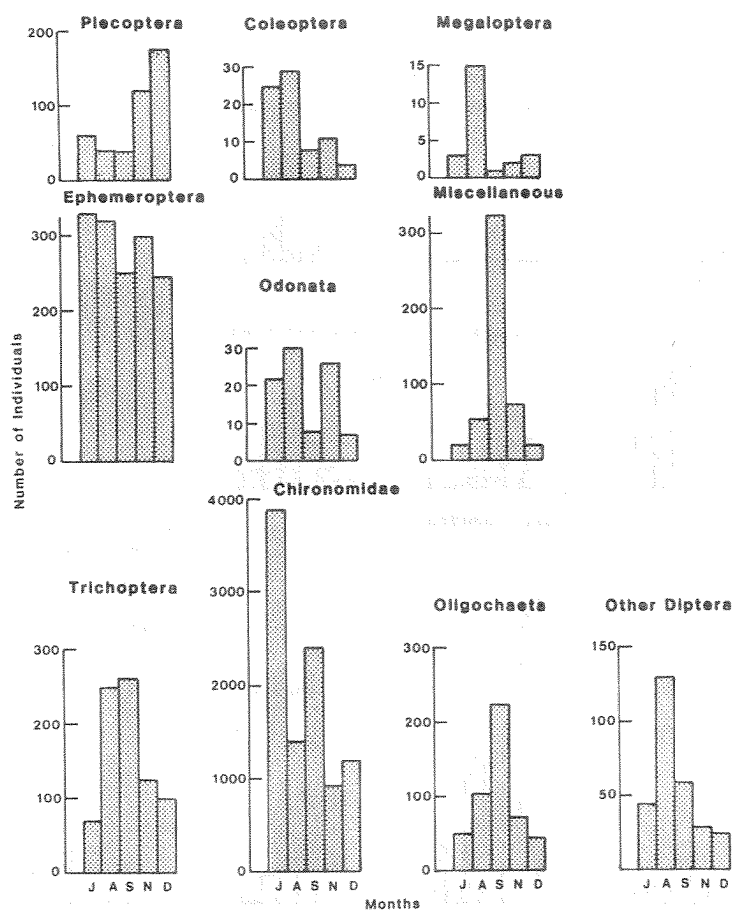


Fig. 4. Monthly variations in the density of the main groups of invertebrates collected from the North River (numbers per 0.05 m<sup>2</sup>) 1982.

STUDY OF PERIPHYTON COMMUNITIES IN ACIDIFIED STREAMS IN NOVA SCOTIA

by

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$$253 + \dots + 1000 = 125175 \quad \text{and} \quad 253 + 254 + \dots + 1000 = 375000$$

$$253 + 254 + \dots + 1000 = 375000$$

$$\frac{253 + 1000}{2} \times 748 = 375000$$

$$\frac{253 + 1000}{2} \times 748 = 375000$$

## ABSTRACT

The major objectives of the investigation are to provide a taxonomic basis for knowledge of algal communities and some understanding of the seasonal changes in streams of mean pH values between 4.7 and 6.3. Statistical analyses of these data indicate that the five study rivers were significantly different in terms of their periphyton biomass. However, if the diatom fraction of each periphyton assemblage is grouped according to pH preference, the four with a mean pH below 5.2 can be distinguished from the remaining streams.

## RÉSUMÉ

L'étude décrite ci-dessous avait comme objectif principal de fournir une base taxonomique à la connaissance des communautés algales et de mieux comprendre les changements saisonniers qui se produisent dans les cours d'eau de pH moyen compris entre 4,7 et 6,3. À l'analyse statistique des données, on constate que les cinq rivières étudiées diffèrent de façon significative sous le rapport de la biomasse périphtonique. Cependant, si l'on groupe par ordre de préférence de pH la fraction diatomée de chaque groupe de périphton, on peut séparer des autres les quatre cours d'eau qui ont un pH moyen inférieur à 5,2.

## INTRODUCTION

Freshwater systems in southern and western Nova Scotia are considered to be potentially susceptible to pH change due to the absence of limestone and other buffering bedrocks. These systems are also subjected to the most intense acid precipitation in Canada (Anon. 1981). However, a large proportion of lakes and streams in these regions of Nova Scotia are dystrophic, originating in acid bogs and forests. Consequently, many streams have an indigenous mean pH around or below the pH 4.7 reported by Underwood (1981) for precipitation falling on the area (Farmer et al. 1980). Thus, this region is subjected to increases in acidity from both anthropogenic and natural sources.

The present study forms part of a large acid rain program aimed primarily at investigating possible effects of increases in the acidity of Nova Scotian streams on their salmonid stocks. Although most lotic ecosystems are heterotrophic, considerable autotrophic production can occur in shallow streams flowing over stable bedrock (Hill and Webster 1982). The major objectives of the investigation are to provide a taxonomic basis for knowledge of algal communities and to understand something of the seasonal changes in streams of different mean pH, thus enabling an assessment of the effect of pH on lotic algal communities.

## MATERIALS AND METHODS

Four of the study rivers were situated in Nova Scotia, Canada: the Bear River (44°35' N 65°40' W), the La Have River (44°40' N 64°50' W), a tributary of the Medway River (44°15' N 64°55' W), and the Westfield River (44°25' N 65°00' W). A New Brunswick river, the Digdeguash (44°25' N 67°12' W), was included to extend the pH range studied to above 6. All five rivers are shallow (maximum depth 1.25 m) and flow over stable bedrock composed of flat stones (approximately 0.1 m<sup>2</sup>) interspersed with large boulders (1 m<sup>3</sup>). The four rivers in Nova Scotia are situated on non-buffering bedrock. The Bear River is located in a region of white rock formation consisting of quartzite, slate, siltstone, rhyolite, tuff and basalt; the La Have River is on the goldville formation, consisting primarily of greywacke and slate. The Westfield River and Medway tributary are on the Halifax formation comprising principally slate and siltstone (Dept. of Mines and Energy, Nova Scotia, Geological Map, 1979). The Digdeguash is situated on a bedrock of greywacke, slate, siltstone, sandstone, conglomerate and limestone (Dept. of Natural Resources, Geological Map No. N.R. 1, 1968).

The determination of pH, periphyton and major invertebrate taxa continued for a full year from August 1981. Samples were collected monthly during the late fall and winter and at biweekly intervals during the remainder of the year. Water depth of the rivers was recorded from a depth marker pole positioned in each river for the duration of the survey. Temperature was measured directly with a mercury thermometer. An Orion Research pH meter Model 407A and combination probe were used to record pH directly at each site to a precision of 0.05 pH units.

The epipellic algal community was surveyed, using the methodology of Blinn et al. (1980). An algal sample was removed from a 2 cm<sup>2</sup> area of rock with the use of a stiff bristled nylon brush and preserved in a vial with Lugol's iodine. The cleaned area of rock was etched after each collection to avoid sampling the same area twice. The epiphytic algal samples were removed from a 5-cm distal section of an aquatic macrophyte species regularly present in each river. The sampling techniques and laboratory methods of Moss (1981) were employed for the analysis of epiphytic algae. The macrophyte samples were drained in the field and placed separately in vials. In the laboratory, filtered water was added and the plants were shaken vigorously for 3 min and the supernatant retained. This procedure was repeated until the water remained clear after shaking. The volume of water was recorded and the algae preserved with Lugol's iodine prior to counting. In each case three replicate samples were collected of which 20 subsamples were counted, using an AO microscope and X40 objective.

## RESULTS AND DISCUSSION

A total of 83 algal taxa were identified in the periphyton of the five study rivers (Appendix 1). All algal taxa within the samples have been recorded, including some species of planktonic origin. Physico-chemical features of the five rivers are recorded in Table 1. The mean seasonal pH values of the Medway tributary, Bear and Westfield Rivers were 4.7, 4.8, and 4.8, respectively. The mean pH's of the less acidic La Have and Digdeguash Rivers were 5.2 and 6.3. The Digdeguash River experienced the same level of acid precipitation as the study rivers in Nova Scotia (Cowling 1982), yet maintains a higher pH due to the influence of buffering bedrocks.

Total number of species recorded in the epiphytic and epipellic communities throughout the year are recorded in Appendices 2-6. Distribution of species number among the four sites has been presented in addition to the total number of algal cells for each river. Data on standing crop, expressed as total number of algae, are presented in Appendices 7-11. Principal components analysis (PCA) of the diatom species data was performed as a means of identifying major parameters (such as pH) involved in influencing the character of the periphytic communities. The PCA was based on a correlation matrix and up to 17 Eigenvectors were generated. Initially, ordinations were produced for the epipellic and epiphytic data sets to investigate the seasonal cluster relationships of one river to another. Subsequently, PCA's were performed on the two algal data sets for each individual river to assess seasonal clustering within the river. The first five principal components combined almost equally to provide 50% of the total variation within the data. Eight parameters were required to obtain 70% of the total variability. Thus, ordinations were of limited value in identifying the variables influencing the lotic systems and have not been included.

The chi<sup>2</sup> statistic was also employed to assess the relationships between rivers. Contingency tables were constructed for each pair of rivers, based on the quantitative data of the 16



major species of the periphytic community. These statistical tests indicated that all the rivers were significantly different in terms of their periphyton biomass.

The algal assemblages of both the epipellic and epiphytic communities for the five study rivers were principally composed of diatoms. These diatom species have been grouped according to pH preference as obtained from the literature (Lowe 1974; Patrick and Reimer 1966, 1972). The data for each river have been replotted in the form of a histogram with the diatom species divided into the following categories: acidobiontic, acidophilous, alkaliphilous, and indifferent. The diatom assemblages of the four rivers in Nova Scotia show a similar seasonal distribution of pH groupings (Fig. 1, 2). The majority of diatoms fall into an acidophilous category. The only acidobiont recorded was *Actinella punctata* var. *punctata* (Lewis) which occurred in January in the Westfield River. The Digdeguash, with a pH consistently above 6, showed a higher population of alkaliphilous diatom species in both the epiphytic and epipellic communities, particularly during the fall and spring blooms. The grouping of diatoms according to pH preference was the only method of presentation which separated the acidic rivers of Nova Scotia from the more alkaline Digdeguash.

As previously discussed, many factors are involved in determining the character of both the epipellic and epiphytic communities. Macrophyte host type has been reported as modifying epiphyte assemblages. Moss (1981) found that the influence of the host macrophyte on the epiphytic community was greater in dystrophic water. Use of the  $^{14}\text{C}$  method for periphyton production estimates in the study rivers (Elner and Johnston, unpublished data) gave rates lower than those reported for many rivers (McConnell and Sigler 1959; Hill and Webster 1982), indicating that the host type may be an important variable. However, due to seasonal heterogeneity in macrophyte production between rivers, it was not always possible to sample the same species of host.

Blinn et al. (1980) considered current velocity, light energy and nutrient budget to be more important than substrate type in determining the epipelon. The data from our study support Blinn's concept. The highest epipellic biomass (Appendices 2-6) occurred in the Medway tributary which was also the shallowest river with the greatest light penetration to the epipelon.

From the histograms (Fig. 1 and 2) it is evident that periphytic species composition can be influenced by pH. However, in the Westfield study area where the pH range is limited, the seasonal influences of current velocity, light energy, nutrient budget and macrophyte diversity predominate in determining the characteristics of the periphytic communities.

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Table 1. Physico-chemical data from five sample rivers.

Date	Depth cm	Temperature °C	pH
BEAR RIVER			
29.07.81	33	17.0	
27.08.81		14.0	5.00
17.09.81			
05.10.81	34	10.0	
20.10.81	23	9.0	4.75
02.11.81	46	7.0	4.8
01.12.81	36	1.0	5.3
07.01.82	66	0.1	4.8
02.02.82		0.1	4.8
04.03.82		0	4.8
05.04.82	65	2	4.6
27.04.82	66	8.5	4.8
12.05.82	31	12	4.7
01.06.82	22	18	5.2
23.06.82	20	17	4.9
07.07.82	17	21	4.6
21.07.82	23	20	5.2

## TRIBUTARY OF THE MEDWAY

29.07.81	21	15.0	
27.08.81	2	18.0	5.0
17.09.81	7	8.0	4.7
05.10.81	8	10.0	4.8
20.10.81	27	8.0	4.9
02.11.81	13	5.0	4.6
01.12.81	8	1.0	5.2
07.01.82	7	1.0	4.9
02.02.82		0.1	4.6
04.03.82		0.1	4.2
05.04.82	31	0.1	4.7
27.04.82	50	6.0	4.6
12.05.82	8	9.0	4.7
01.06.82	7	13.0	4.8
23.06.82	115	15.0	4.5
07.07.82	2.5	22.0	4.5
21.07.82	20	21.0	4.7

## DIGDEGUASH RIVER

29.07.81	10	15.0	6.3
27.08.81		14.0	
17.09.81			
05.10.81			
20.10.81	34	7.0	6.3
02.11.81	30	2.0	6.2
01.12.81	37	0.1	6.2
07.01.82			6.3
02.02.82			
04.03.82		0.1	5.8
05.04.82	42	1.0	6.2
27.04.82	40	10.0	6.2
12.05.82	11	9.0	6.2
01.06.82	-10	21.0	6.6
23.06.82	3	16.0	6.5
07.07.82	9	19.0	6.6
21.07.82	-15	27.0	7.0

Table 1. continued

Date	Depth cm	Temperature °C	pH
LA HAVE RIVER			
29.07.81	10.5	18.0	
27.08.81		14.0	5.0
17.09.81	4	8.0	
05.10.81	27	11.0	
20.10.81	33	9.0	5.6
02.11.81	34	8.0	5.5
01.12.81	36	1.0	5.1
07.01.82	63	0.1	5.5
02.02.82		0.1	5.4
04.03.82		0.1	4.6
05.04.82	55	3.0	4.8
27.04.82	62	8.0	5.3
12.05.82	25	12.5	5.4
01.06.82	12	16.5	5.4
23.06.82	22	15	5.1
07.07.82	30	22	5.3
21.07.82	10	22	5.3

## WESTFIELD RIVER

29.07.81	22	22.3	
27.08.81	7	20.0	
17.09.81	8	18.0	5.2
05.10.81	16	10.5	5.2
20.10.81	27	9.0	5.4
02.11.81	46	7.0	4.8
01.12.81		1.0	4.8
07.01.82		1.0	4.7
02.02.82		1.0	4.6
04.03.82	8	1.0	4.5
05.04.82		1.0	4.4
27.04.82	110	9.0	4.6
12.05.82	58	12.0	4.7
01.06.82	22	18.0	4.8
23.06.82	45	19.0	4.8
07.07.82	20	22.0	4.9
21.07.82	22	22.0	4.9

## Appendix 1. List of algal taxa recorded during the study.

## DIATOMS

## PENNATES

Achnanthes flexella  
A. minutissima v. minutissima  
Actinella punctata v. punctata  
A. exigua v. constricta  
Anomoeoneis foliis v. foliis  
Asterionella formosa  
A. ralfsii v. ralfsii  
Caloneis clevei  
Cocconeis placentula v. lineata  
Cymbella aspera  
C. lunata v. lunata  
C. minuta  
C. microcephala  
Denticula elegans  
Diatoma anceps v. anceps  
D. hiemale v. mesodon  
Eunotia arcus v. bidens  
E. bidentula v. bidentula  
E. curvata v. curvata  
E. diodon v. diodon  
E. incisa  
E. meisteri v. meisteri  
E. pectinalis v. pectinalis  
E. pectinalis v. ventricosa  
E. serra v. diadema  
E. serra v. serra  
E. sudetica  
E. tautoniensis v. tautoniensis  
E. tenella v. tenella  
E. trinacria  
E. vanheurckii v. intermedia  
E. vanheurckii v. vanheurckii  
E. a  
E. b  
Fragilaria construens v. binodis  
F. construens v. subsalina  
F. construens v. venter  
Frustulia rhomboides v. capitata  
F. rhomboides v. capitata b  
F. rhomboides v. rhomboides  
F. rhomboides v. saxonica  
Gomphonema acuminatum v. acuminatum  
G. carolinense  
G. parvulum v. parvulum  
G. truncatum v. truncatum  
Navicula minima v. minima  
N. radiosa v. radiosa  
N. seminulum v. seminulum

N. viridula v. linearis  
N. a  
N. b  
Nitzschea palea  
Pinnularia abaujensis  
P. abaujensis v. subundulata  
Stauroneis anceps v. gracillia  
Surirella linearis v. constricta  
S. linearis v. linearis  
Synedra fasciculata  
S. rumpens  
S. ulna  
S. ulna v. biceps  
Tabellaria fenestrata  
T. flocculosa  
 Unidentified Pennates

## CENTRICS

Cyclotella kutz  
Melosira distans  
M. granulata  
M. italica

## BLUEGREENS

## CHLOROPHYTA

Actinastrum  
Ankistrodesmus  
Crucigenia rectangularis  
Distyosphaerium  
Pediastrum  
Scenedesmus obliquos  
S. quadricauda  
Selenastrum pandorina  
S. westii

## FILAMENTOUS CHLOROPHYTA

## DESMIDS

Closterium calosporum  
Closterium moniliferum  
Cosmarium  
Staurastrum hexacerum  
S. orbiculare

Appendix 2. Total number of species in the epipellic and epiphytic communities of the tributary of the Medway River.

Date	Epipellic				Epiphytic			
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Shady riffle	Sunny riffle	Shady pool	Sunny pool
28.07.81	3	11	13	11				
28.08.81	14	11	12	11				
16.09.81	8	9	13	12	5	10	4	
30.09.81	12	11	7	10	1	10		
19.10.81	10	15	4	11	10	15	4	11
01.11.81	8	7	7	5	8	7	7	5
30.11.81	15	16	13	11	15	16	13	11
07.01.82	5	1	9	13	5	1	9	13
02.02.82	8	7		6				
04.03.82			5					
01.04.82		6	10	10		7	14	3
27.04.82	12	11		14	9	7		7
12.05.82	6	7	13	15			7	10
01.06.82	8		6	9	7	1	6	
26.06.82	15	11	7	11	5		10	
08.07.82	5	6	6	3	6	7	9	4
21.07.82	10	5	11	7	8	7	6	6

Appendix 3. Total number of species in the epipellic and epiphytic communities of the Bear River.

Date	Epipellic				Epiphytic			
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Shady riffle	Sunny riffle	Shady pool	Sunny pool
28.07.81	4	12	11	6	13	14	16	9
28.08.81	2	5	5	12	6	15	10	14
16.09.81	7	6	5	5	8	6	8	7
30.09.81	4	9	10	8	10	12	9	11
19.10.81	3	4	4	2	9	4	4	10
01.11.81	11	1	3	4	5	10	9	13
30.11.81	2	2	1	5	7	11	8	13
07.01.82	1		11	3	7			4
02.02.82			4		6			7
04.03.82		2		3				
01.04.82	6		6	5	13		6	9
27.04.82	5	5			11	9		
12.05.82	9	3	11	12	10	8	9	14
01.06.82	5	6	11	8	8	4	5	10
26.06.82	7	4	6	3	8	8	6	9
08.07.82	3	4	2	8	5	9	10	10
21.07.82	4	2	10	7	10	3	4	9

Appendix 4. Total numbers of species in the epipellic and epiphytic communities of the Westfield River.

Date	Epipellic				Epiphytic			
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Shady riffle	Sunny riffle	Shady pool	Sunny pool
28.07.81	9	9	10	15				
28.08.81	24	18	12	20	16	7		11
16.09.81	18	25	14	16				
30.09.81	5	14	19	16				
19.10.81	8	11	14	8			1	4
01.11.81		8	8				1	1
30.11.81	9	9	5	20				
07.01.82		5	5	3			1	4
02.02.82	7		9					
04.03.82	4	10	9	7				
01.04.82		5	16	8		4		
27.04.82		8	8			2		
12.05.82		11	14	18				14
01.06.82	13	7	13	6				2
26.06.82	12	14	9	9			3	4
08.07.82	13	6	13	19			6	5
21.07.82	18	12	13	11			9	5

Appendix 5. Total number of species in the epipellic and epiphytic communities of the La Have River.

Date	Epipellic				Epiphytic			
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Shady riffle	Sunny riffle	Shady pool	Sunny pool
28.07.81	15	10	3	7				
28.08.81	8	4	7	15		6	6	14
16.09.81	8	11	5	14	11	3	2	3
30.09.81	4	8	7	10				
19.10.81	3	6	2	4		5		
01.11.81	3	11	17	3	6	4	4	
30.11.81	10		3	2	7		5	
07.01.82	2		4		14	1	4	
02.02.82	6							
04.03.82				2				
01.04.82	9	3			4	1		
27.04.82	4			6	7			
12.05.82	9	16	2	11	9	10		
01.06.82	6	5	7	5	10	5	9	6
26.06.82	6	4	5	9		7	3	1
08.07.82	10	7	15	17	3	2	4	2
21.07.82	12	10	15	12	5	2	3	2

Appendix 6. Total number of species in the epipellic and epiphytic communities of the Digdeguash River.

Date	Epipellic				Epiphytic			
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Shady riffle	Sunny riffle	Shady pool	Sunny pool
28.07.81	12	3	5	2				
28.08.81	1	7		2				
16.09.81	10	5	30	29	9	4	13	
30.09.81	2	3	3	10	2	1	1	
19.10.81	3	4	4	2				6
01.11.81	11	1	3	4	9	2	6	4
30.11.81	2	2	1	5			11	6
07.01.82	1		11	3				
02.02.82								
04.03.82								
01.04.82	10	5			2	3		
27.04.82	4	1			10			
12.05.82	3	3	8	5	8			9
01.06.82	5	6	3	10	2	7		5
26.06.82	21	6	11	20	16		16	
08.07.82	10	7	15	17	5	5	7	13
21.07.82	12	10	15	12		3	11	7

Appendix 7. Standing crop for the epipellic and epiphytic communities of the tributary of the Medway River.

Date	Epipellic (algae $m^{-2} \times 10^8$ )					Epiphytic algae g (dry wt; macrophyte) $^{-1} \times 10^3$				
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total
28.07.81	36	19	1 494	594	1 006					
28.08.81	5 508	1 117	4 707	657	2 997					
16.09.81	963	459	837	1 251	877	4 898	710	120		1 909
30.09.81	648	4 644	288	5 202	2 695	90	779			434
19.10.81	12 312	23 292	108	4 185	9 974	3 921				980
01.11.81	981	387	495	835	675	710	1 000	30 760	1 499	11 325
30.11.81	2 628	4 149	1 116	1 206	2 279			866	685	775
07.01.82	1 629	18	15 930	1 575	4 788				275	68
02.02.82	931	1 125		468	631					
04.03.82			1 255		313					
01.04.82		690	15 316	18 978	8 746		2 492	1 373	384	1 416
27.04.82	>40 000	>40 000		>40 000	>40 000	3 659	2 409		3 835	3 301
12.05.82	>40 000	>40 000	>40 000	9 945	>40 000			12 591	>40 000	>40 000
01.06.82	>40 000		>40 000	>40 000	>40 000	869	17	1 034		480
26.06.82	1 741	1 546	340	781	115	1 113		2 709		1 911
08.07.82	225	202	111	31	58	137	606	717	220	420
21.07.82	528	333	320	123	122	530	518	173	461	420

Appendix 8. Standing crop estimates for the epipellic and epiphytic communities of the Bear River.

Date	Epipellic					Epiphytic				
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total
28.07.81	477	845	738	135	548	631	166	483	145	318
28.08.81	27	216	198	180	155	55	1 287	201	1 840	431
16.09.81	99	198	189	63	137	1 064	300	1 099	359	705
30.09.81	981	693	486	513	668	402	2 038	454	621	878
19.10.81	90	477	36	18	155	989	206	165	725	521
01.11.81	657	99	54	306	279	1 137	1 803	664	775	1 094
30.11.81	18	18	9	81	31	289	482	644	839	563
07.01.82	2 682		361	171	1 282	158			23	90
02.02.82			274		68	442			604	523
04.03.82		22		90	28					
01.04.82	401		298	2 551	812	183		33	208	141
27.04.82	3 612	>40 000			>40 000	215	281			248
12.05.82	268	39	294	>40 000	>40 000	938	244	287	2 144	903
01.06.82	97	122	627	78	231	861	240	282	438	455
26.06.82	168	60	191	42	115	227	313	392	918	392
08.07.82	26	30	17	161	58	145	457	571	960	683
21.07.82	35	99	186	169	122	286	43	62	257	162

Appendix 9. Standing crop estimates for the epipellic and epiphytic communities of the Westfield River.

Date	Epipellic					Epiphytic				
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total
28.07.81	288	1 422	1 350	1 251	1 077					
28.08.81	5 013	1 575	2 547	2 259	2 878	35 705	670		5 375	2 937
16.09.81	1 665	8 676	2 358	3 366	4 016				6 727	1 681
30.09.81	315	1 611	4 365	1 377	1 917					
19.10.81	1 368	1 314	802	639	1 030			225	1 093	659
01.11.81		126	153		69			19	236	137
30.11.81	1 773	298	162	1 961	1 048					
07.01.82		54	162	36	63			4	29	17
02.02.82	859		51		302					
04.03.82	1 498	1 917	472	697	1 146					
01.04.82		2 525	11 509	5 993	5 006		372			90
27.04.82		>40 000	1 336		>40 000		66			16
12.05.82		7 323	3 976	782	3 020				4 163	1 040
01.06.82	>40 000	4 030	>40 000	1 335	>40 000				124	31
26.06.82	1 357	944	368	137	701			165	632	398
08.07.82	1 500	986	867	731	1 021			196	191	193
21.07.82	439	830	754	433	614			333	38	185

Appendix 10. Standing crop estimates for the epipellic and epiphytic communities of the La Have River.

Date	Epipellic					Epiphytic				
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total
28.07.81	729	225	90	198	310					
28.08.81	189	36	99	369	173		52	156	553	253
16.09.81	171	360	3 807	618	1 239	401	548	83	444	369
30.09.81	81	324	216	387	252		80			20
19.10.81	54	149	72	243	129	298	438	76		270
01.11.81	72	270	693	36	267	347		704		524
30.11.81	378		72	126	144	297			553	425
07.01.82	27		126		76	445	43	35		174
02.02.82	486				121					
04.03.82				175	43				1 683	420
01.04.82	121	87			52	200	59			129
27.04.82	>40 000			106	>40 000	115				28
12.05.82	671	697	31	1 572	742	239	726			482
01.06.82	776	948	129	849	675	1 121	185	1 620	425	837
26.06.82	61	123	213	265	163		63	48	75	62
08.07.82	307	553	83	152	273	36	36	145	106	80
21.07.82	483	1 176	326	218	550	102	77	185	187	137

Appendix 11. Standing crop estimates for the epipellic and epiphytic communities of the Digdeguash River.

Date	Epipellic					Epiphytic				
	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total	Shady riffle	Sunny riffle	Shady pool	Sunny pool	Total
28.07.81	819	90	387	36	333					
28.08.81	9	135		36	45					
16.09.81	297	1 620	2 034	5 040	2 249	490	125	995		248
30.09.81	117	828	72	441	364	45	13	2		15
19.10.81			1 305	2 106	852				149	37
01.11.81	4 344		432	1 674	1 613	11 392	257	184	983	3 204
30.11.81			81	621	351			115	2 080	548
07.01.82	1 917				479					
02.02.82										
04.03.82										
01.04.82	524	124			162	67	95			81
27.04.82	94	>40 000			>40 000	301				75
12.05.82	66	21	574	107	192	176			96	91
01.06.82	58	132	265	271	181	108	315		144	141
26.06.82	2 370	91	368	3 542	1 592	238		940		589
08.07.82	353	547	706	2 401	1 001	67	80	296	2 788	807
21.07.82	426	172	752	527	469		66	328	238	225



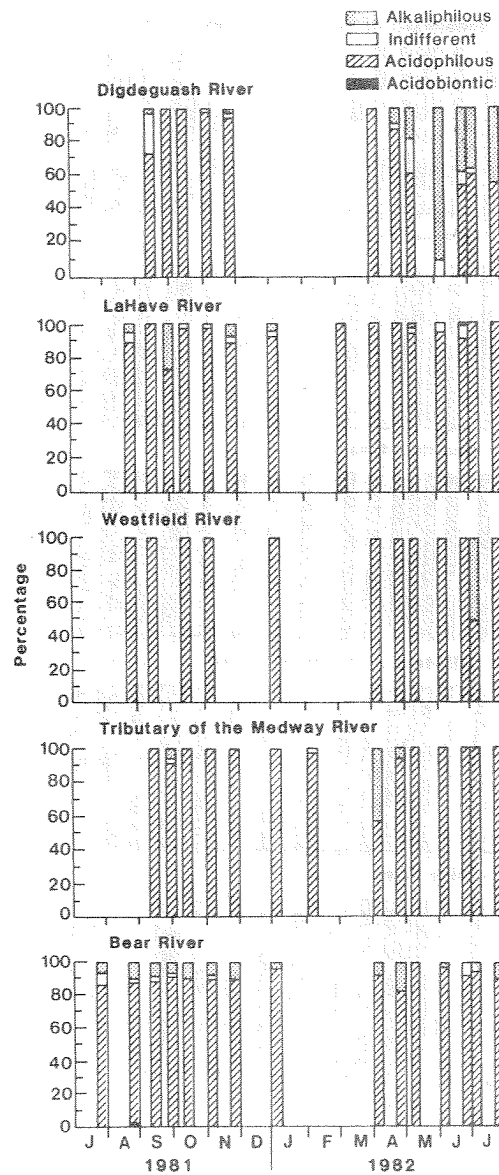


Fig. 1. Diatom pH categories for the epiphytic community of the five rivers.

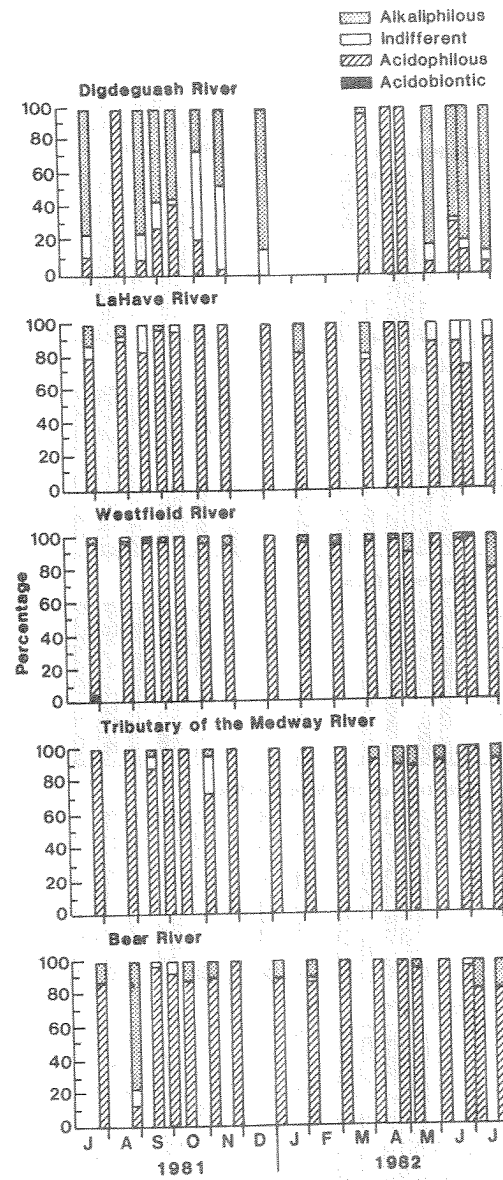


Fig. 2. Diatom pH categories for the epipelton of the five rivers.

A STUDY OF SEX HORMONE PRODUCTION, SEXUAL MATURATION,  
AND REPRODUCTION IN ATLANTIC SALMON (SALMO SALAR) HELD IN THE  
WESTFIELD AND MEDWAY RIVERS, QUEENS CO., NOVA SCOTIA

by

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## ABSTRACT

Two-year-old Atlantic salmon (*Salmo salar*) of the same stock were held in cages in the low pH Westfield River and the higher pH Medway River for comparison. Both groups of fish were fed daily the same diet. The Medway River fish ate better, gained more weight, produced larger eggs, and had lower mortality. The blood sex hormone levels in the Medway River fish were normal but there were indications that the hormone levels in the Westfield River fish were not.

## RÉSUMÉ

Des saumons atlantiques (*Salmo salar*) de 2 ans d'un même stock ont été maintenus dans des cages placées dans la rivière Westfield à bas pH et, pour comparaison, dans la rivière Medway à pH plus élevé. Les deux groupes ont reçu quotidiennement la même ration. Les poissons de la rivière Medway mangèrent d'un meilleur appétit, augmentèrent davantage de poids, produisirent des oeufs plus gros et subirent des mortalités moindres. Les niveaux d'hormones sexuelles du sang des poissons de la rivière Medway étaient normaux, mais on a des indications que ceux des poissons de la rivière Westfield ne l'étaient pas.

## INTRODUCTION

In 1981 we found that the blood levels of the two principal androgens, 11-ketotestosterone and testosterone, were much lower in sexually mature male Atlantic salmon (*Salmo salar*) sampled on their spawning migration in the Westfield River (pH 4.7) than in salmon from the Medway River (pH 5.6) sampled at the same time (Freeman 1982). Testosterone and 11-ketotestosterone increase in concentration in the blood of normal male Atlantic salmon during sexual maturation and peak at or near functional maturity (Idler et al. 1971; Sangalang and Freeman 1974; Hunt et al. 1982). The direct relationship between androgen production and maintenance of spermatogenesis in vertebrates including fish is well known (Ozon 1972). Therefore, we investigated the relationship between blood sex hormone levels and sexual maturation to determine if the Atlantic salmon in the low pH Westfield River have reproduction problems. The weight gain from early maturity to functional sexual maturity of the Westfield River salmon was also compared with similar salmon in the less acidic Medway River.

In 1982, 54 maturing Atlantic salmon, equal numbers of each sex, were placed in a cage in the Westfield River. The same number of Atlantic salmon were held in a cage in the higher pH Medway River for comparison. The Westfield River is a small tributary of the Medway River which enters the Medway from a northeasterly direction approximately 24.1 km upstream from the Medway River at Greenfield, Queens Co., Nova Scotia.

## MATERIALS AND METHODS

Two-year-old Atlantic salmon of Medway River origin were purchased from the Cape Breton Marine Farm, Baddeck, Nova Scotia. These salmon were tagged at Baddeck with numbered plastic tags (Eisner and Ritter 1979). The sex of the fish was determined by analysis of blood plasma by radioimmunoassay, using 11-ketotestosterone antiserum as described by Sangalang et al. (1978). The salmon were transported to a floating cage anchored in the Medway River at Greenfield, Nova Scotia, on July 13 and 15, 1982, where they were held and fed daily 6.35 mm trout grower pellets purchased from Co-Op Atlantic, New Minas, Nova Scotia, until July 28, 1982, when the fish were divided into two groups of equal numbers containing equal numbers of each sex. One group was held in the cage in the Medway River and the other was held in a similar cage in the Westfield River.

The fish cages were constructed by suspending a 3.18-cm mesh nylon net (3.66 x 2.44 x 1.83 m deep) from the open center of a rectangular wooden platform buoyed by 12.7-cm thick styrofoam. The platform had outside measurements of 4.87 x 6.10 m. The open section was fenced at the top of the platform with 0.61-m high chicken wire (2.5-cm mesh). The fenced opening which provided access to the fish was covered with chicken wire secured in a "snug-fitting" but removable wooden frame. The suspended cage was protected from predators and debris by a 2.5-cm mesh chicken wire enclosure suspended from the outer edge of the wooden platform of the cage so that it was a foot outside the nylon netting.

The fish at both locations continued to be fed daily the trout grower pellets. On August 23 and 24, 1982, the numbers of fish in each cage were reduced to 42. Starting on August 23, 1982, on the Westfield River and August 24, 1982, on the Medway River and then at 2- to 3-wk intervals the fish were removed from the cages, anaesthetized with MS222, weighed and approximately 1 mL of blood was collected in heparinized syringes by caudal venopuncture. Hematocrits were determined. Blood plasma were separated by centrifuging and stored at -40°C prior to subsequent analysis for 11-ketotestosterone and testosterone by radioimmunoassay (Sangalang and Freeman 1977). The pH and temperature of the river waters were determined at each sampling period.

At spawning time, the fish were anaesthetized, weighed, spawned, and sampled for blood. The mean diameter of the eggs of each female was determined and the packed cell volume (spermocrit) of sperm was determined, using glass capillaries and a centrifuge as for hematocrits.

The eggs from Medway and Westfield fish were fertilized with milt from fish from their respective groups. The eggs were incubated at the Mersey hatchery and the mortality of eggs to the eyed stage was determined.

## RESULTS AND DISCUSSION

pH profiles of the Medway and Westfield River waters at the time of sampling are shown in Fig. 1.

The pH of the Medway River water was higher than that of the Westfield River, with the pH ranging from 5.4 to 6.15 on the Medway and 5.1 to 5.28 on the Westfield. The pH of the water in the Westfield River during the present study period from August to November 1982 was considerably higher than at the time of sampling the wild Atlantic salmon in November 1981 when the pH was 4.7. The higher pH in 1982 was possibly due to very little rainfall during the fall of 1982. This caused lower water levels and a higher water pH in the Westfield River. In the fall of 1981 there was much rainfall resulting in high water levels in the Westfield River and, presumably, contributing to the low pH of 4.7. The recorded water temperatures (Fig. 2) are not the mean temperatures for the day but the temperature at the time of sampling. The Westfield River water temperatures were generally taken at 7:30 to 8:00 A.M. The water temperature in the Westfield fluctuated more than the Medway water temperatures as it was very shallow and therefore the water temperature early in the morning was nearer the air temperature and generally lower than in the Medway whose water temperature fluctuated only slightly due to its large volume of water flow.

At the beginning of the experiment, the mean body weights ( $\pm$ S.D.) of Westfield River and Medway River salmon were 759.1 ( $\pm$ 321.1) and 893.9 ( $\pm$ 354.4) g, respectively (Table 1). The mean body weights of the Medway River fish increased to 1088.1 ( $\pm$ 282.9) g at spawning time (at 70 d) while the mean body weight of the Westfield River salmon increased to 832.0 ( $\pm$ 268.2) over the same period of time (Table 1). This represents an average increase in body weight of 21.7 and 9.60% for the Medway and Westfield River salmon, respectively. Twelve days

after spawning, (at 92 d) the mean body weight of the Medway River salmon was 930.1 ( $\pm 264.5$ ) g while that of the Westfield River salmon was 761.9 ( $\pm 246.9$ ) g. This was an average mean body weight gain of 4.05% for the Medway and only a 0.37% average weight gain for Westfield fish.

The greater weight gain of the Medway fish is explained by the Medway River fish eating well when fed daily. The Westfield fish were not as active and did not eat well although they were fed daily the same diet as the Medway River fish. Their lack of appetite could have been due to the conditions of the water in the Westfield River, suggesting that it was not as suitable for holding salmon as the Medway River waters.

On November 24, 1982, 1 d after the last blood sampling, the Westfield River salmon were moved back to the Medway River where they are now being held. All salmon are being fed daily. The Medway fish still feed better than the Westfield fish. The fish are currently under observation to determine whether the effects of the Westfield River waters are long lasting.

The salmon in both rivers became sexually mature during the same period of time (November 1-15): 17 Medway males and 11 Westfield males ripened; 13 females ripened in each of the two rivers. The egg mortality to the eyed stage for Westfield eggs was 90.9% compared to 59.3% for eggs from the Medway River fish. The mean egg diameters for the Medway and Westfield River salmon were 5.77 and 5.46 mm respectively, indicating that the egg volume of the Medway River fish was 16% greater than those taken from the Westfield River fish. It is not known whether the smaller mean volume of the Westfield River fish eggs is due to the fish being smaller in size, for at the beginning of the experiment the Westfield River fish weighed 15% less than the Medway River fish, or whether it is due to other factors. Larson and Pickova (1978) found a positive correlation between female weight and egg size in all age classes of Atlantic salmon in a study carried out on three river stocks in Sweden. It is known, however, that egg size may be influenced by diet and water temperature (Trever Goff, personal observation). Glebe et al. (1979) found that there was no significant correlation between egg size and survival; therefore it would appear that the high mortality of the Westfield River salmon eggs may not be associated with size.

The packed cell volume of the milt from Medway River fish ranged from 18.0-48.8% (mean  $31.7 \pm 8.4$ ). The packed cell volume of the Westfield River fish ranged from 13.0-61.9% (mean  $40.8 \pm 12.8$ ). At 95% confidence levels (F-test) the two groups are different but we do not know what this implies.

The mean concentrations of 11-ketotestosterone in the blood of both the Medway and Westfield River male salmon increased as the fish matured sexually (Fig. 3), agreeing with what was found in other investigations with maturing salmonids (Idler et al. 1971; Sangalang and Freeman 1974; Hunt et al. 1982). The levels of this androgen peaked at or near functional maturity. The peak values ( $\bar{X} \pm S.D.$ ) of 11-ketotestosterone for Medway and Westfield River fish were  $62.0 \pm 20.1$  ng/mL and  $40.3 \pm 25.9$  ng/mL, respectively (Table 2). These hormone levels are within the range reported by Hunt et al. (1982) who found 11-ketotestosterone peak levels to range from 2.5-7.0 g/100 mL (i.e. 25 to 70 ng/mL) of plasma in

sexually mature male Atlantic salmon in Scotland also during the months of October and November. The mean levels of 11-ketotestosterone in male salmon from the Westfield and Medway Rivers and 22 wild male salmon sampled at the Coldbrook Hatchery, Coldbrook, Nova Scotia, were approximately the same (ca. 40 ng/mL of plasma) November 2-12, 1982 (Table 2); however, the mean concentration of 11-ketotestosterone in the plasma of the male Westfield salmon decreased sharply on November 23, 1982 (Fig. 3). This level differed from those found in the Medway fish (Fig. 3) and from the data obtained from fish of the same sexual maturity in Scotland by Hunt et al. (1982). The results suggest that the metabolism or utilization of 11-ketotestosterone may be abnormal in the Westfield River even when the pH was slightly over 5 and higher than it was in 1981. In November 1981, we found very little or only a trace of 11-ketotestosterone in the blood of wild Atlantic salmon in the Westfield River which at that time had a pH of 4.7.

Testosterone levels in the blood plasma of sexually maturing male Atlantic salmon in the Medway River increased as the fish matured sexually and peaked at or near functional maturity (Table 2), as in the case for normal salmonids (Idler et al. 1971; Sangalang and Freeman 1974; Hunt et al. 1982). The plasma testosterone levels were also in the normal range for the various stages of sexual maturation. The mean levels of plasma testosterone of the Westfield fish did not show a direct correlation with the approach of functional sexual maturity as normally occurs in salmonids (Idler et al. 1971; Sangalang and Freeman 1974; Hunt et al. 1982). There was no sharp decline in plasma testosterone after functional maturity (Fig. 4) as normally occurs.

A similar elevated plasma level of testosterone after spawning and at the onset of testicular regression was observed in brook trout that were held in trace sublethal toxic levels of cadmium (Sangalang and Freeman 1974), suggesting that the elevated testosterone levels in the male Westfield Atlantic salmon may have been caused by the toxicity of some pollutant associated with the low pH of the river water.

Although the physiological effects of the "abnormal" trend of steroid levels in the Westfield fish appear obscure to us at this time, the results of the comparative studies at the Westfield and Medway Rivers strongly suggest some interference with the normal sexual maturation process in the Atlantic salmon in the Westfield River. This possibly could explain the high mortality (90.9%) of the eggs from the Westfield River.

It became apparent late in our field work that the year 1982 was not a good year for studying the effects of low pH on Atlantic salmon in the Westfield River as there was little rainfall, resulting in low river water and a higher pH than in 1981.

In conclusion, this investigation demonstrates differences between the fish held in the Westfield and those held in the higher pH Medway River. The Medway River fish ate better, gained more weight, produced larger eggs, and had lower egg mortality. Blood sex hormone levels in the Medway fish were normal but there were indications that the hormone levels in the Westfield fish were not.

This study should continue as it is important to establish a definite relationship between the quantity of rainfall, the pH of the river water and the effects of low pH on fish. It is important to establish whether a rainfall would affect the pH of the river drastically, and whether this again would translate to the differences we have observed in 1981 and 1982. It is also important to establish if a gradual deterioration of conditions in the Westfield River would continue to be reflected in the sexual maturation or reproduction process, or whether a "chronic condition" would result in a species adapting to its environment. We think that it is important to continue our efforts to answer the many questions that we recognize remain to be answered in the early phases of our study.

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Table 1. Mean body weights of Atlantic salmon at sampling times.

Time (d)	River	Mean body wt $\pm$ S.D. (g)
0 - (August 23/82)	Westfield	759.1 $\pm$ 321.1 (42 fish)
17	Westfield	765.0 $\pm$ 337.7
38	Westfield	800.3 $\pm$ 308.0
58	Westfield	818.3 $\pm$ 299.8
70	Westfield	832.0 $\pm$ 268.2
92 (after spawning)	Westfield	761.9 $\pm$ 246.9
0 - (August 24/82)	Medway	893.9 $\pm$ 354.4 (42 fish)
16	Medway	957.2 $\pm$ 340.2
37	Medway	959.3 $\pm$ 310.5
57	Medway	999.4 $\pm$ 275.0
70	Medway	1088.1 $\pm$ 282.9
92 (after spawning)	Medway	930.1 $\pm$ 264.5

Table 2. Concentrations of 11-ketotestosterone (11-KT) and testosterone (T) in the blood plasma of male Atlantic salmon (*Salmo salar*) held in the Medway and Westfield Rivers, Nova Scotia, and wild spawning male salmon sampled at the Coldbrook Hatchery, Coldbrook, Nova Scotia.

Sampling period 1981	ng 11-KT/mL X $\pm$ S.D. (S.E.M.)		ng T/mL X $\pm$ S.D. (S.E.M.)	
	Medway	Westfield	Medway	Westfield
Aug. 23-24	5.69 $\pm$ 3.80 (0.21)	8.36 $\pm$ 4.28 (0.43)	3.27 $\pm$ 1.74 (0.10)	2.78 $\pm$ 1.5 (0.15)
Sept. 9-10	7.96 $\pm$ 2.32 (0.12)	11.7 $\pm$ 7.6 (0.59)	8.23 $\pm$ 3.86 (0.21)	19.9 $\pm$ 7.61 (0.69)
Sept. 30	13.1 $\pm$ 6.05 (0.34)	18.7 $\pm$ 10.7 (0.89)	15.6 $\pm$ 10.8 (0.67)	12.2 $\pm$ 3.46 (0.32)
Oct. 20	62.0 $\pm$ 20.1 (0.13)	40.3 $\pm$ 25.9 (0.32)	20.8 $\pm$ 10.9 (0.64)	19.6 $\pm$ 10.3 (0.10)
Nov. 1-15	37.5 $\pm$ 12.7 (0.11)	38.3 $\pm$ 17.6 (0.13)	18.7 $\pm$ 8.55 (0.53)	11.8 $\pm$ 3.97 (0.33)
Nov. 23	29.6 $\pm$ 19.8 (0.14)	7.64 $\pm$ 5.28 (0.59)	13.2 $\pm$ 5.96 (0.37)	20.6 $\pm$ 10.7 (0.22)
	Coldbrook Hatchery		Coldbrook Hatchery	
Nov. 2-12	40.3 $\pm$ 14.7 (0.64)		6.75 $\pm$ 3.71 (0.23)	

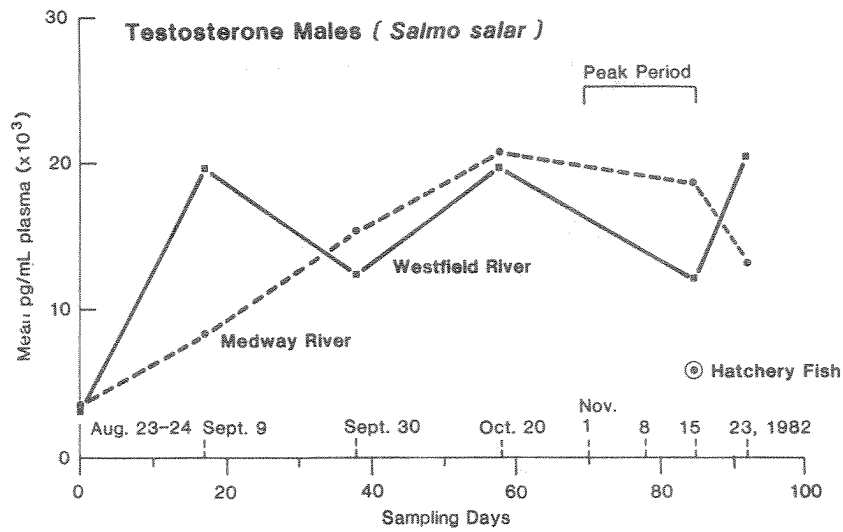


Fig. 1. Water pH of Medway and Westfield Rivers from August 23-24 to November 23, 1982.

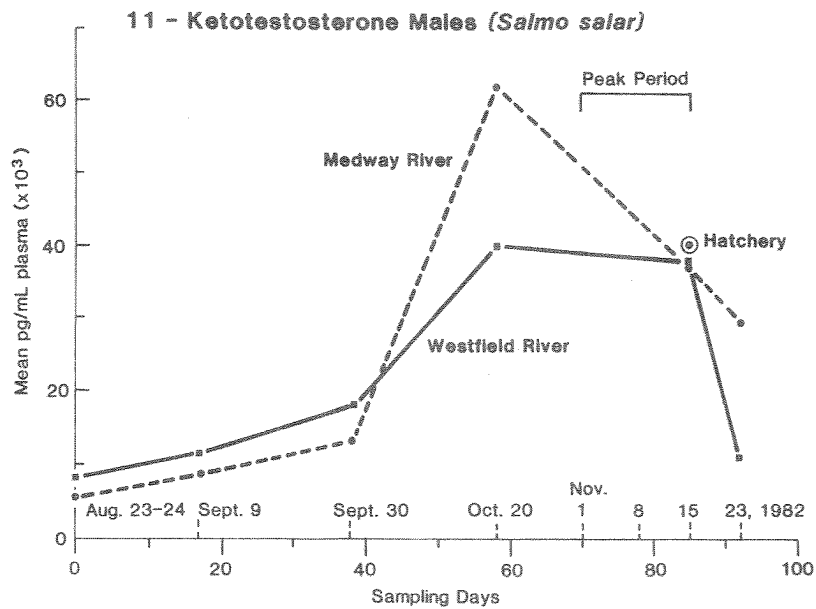


Fig. 2. Water temperatures of Medway and Westfield Rivers from August 23-24 to November 23, 1982.

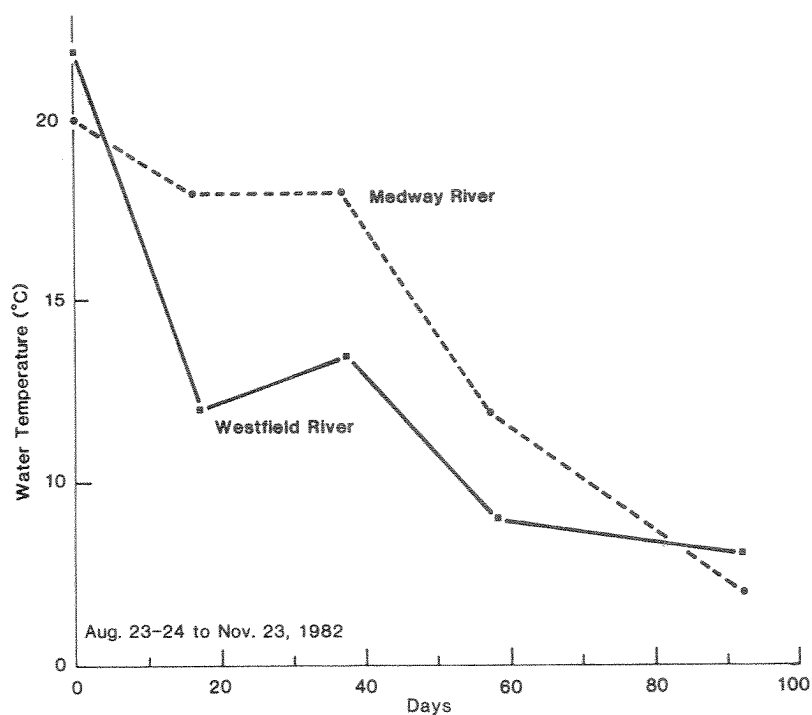


Fig. 3. Blood 11-ketotestosterone levels of male Atlantic salmon (*Salmo salar*) held in the Medway and Westfield Rivers from August 23-24 to November 23, 1982.

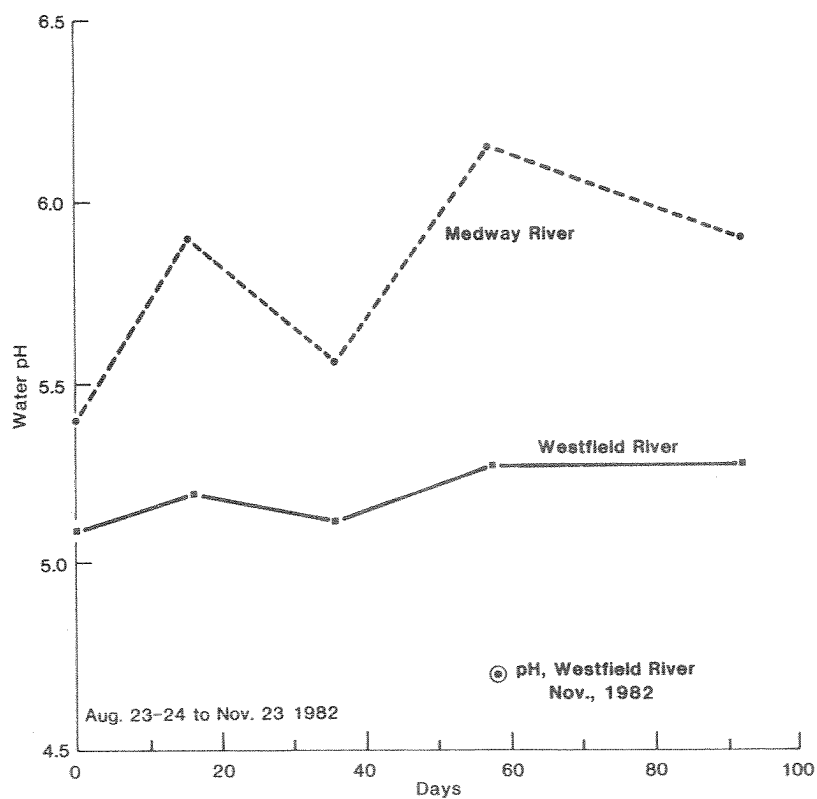


Fig. 4. Blood testosterone levels of male Atlantic salmon (*Salmo Salar*) held in the Medway and Westfield Rivers from August 23-24 to November 23, 1982.



PHYSIOLOGICAL EFFECTS OF LOW pH ON THE SMOLTING PROCESS  
IN ATLANTIC SALMON<sup>1</sup>

by

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<sup>1</sup>A complete account of this work is to be published elsewhere.

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## ABSTRACT

This paper reports on studies conducted between early March and late May to show how low environmental pH affects smolting of *Salmo salar*. Temperatures ranged from 2.3 to 10.0°C and three pH levels were maintained: mean 6.78 (control); mean 4.94, and mean 4.66. There was a high rate of mortality in the low as compared with the intermediate and control pH levels. Growth was reduced in the low pH range. Salinity tolerance (to 35.0 ‰ sea water) was marginally reduced in the intermediate pH group and greatly reduced in the low pH group in comparison with control fish.  $\text{Na}^+\text{K}^+$  ATPase activity was likewise reduced in the intermediate and low pH groups. Plasma  $\text{Na}^+$ ,  $\text{Cl}^-$  and  $\text{Ca}^{++}$  regulation was adversely affected in the intermediate and low pH group. Under the conditions of our experiment, smolting of salmon was inhibited by environmental pH's below 5.

## RÉSUMÉ

Nous décrivons dans le présent article des études menées entre le début de mars et la fin de mai dans le but de démontrer l'influence du pH ambiant sur la smoltification de *Salmo salar*. Les températures ont varié de 2,3 à 10,0°C et trois niveaux de pH ont été utilisés: moyenne de 6,78 (témoins); moyenne de 4,94 et moyenne de 4,66. Les mortalités ont été élevées aux bas pH comparativement aux pH intermédiaires et témoins. Dans la gamme des bas pH, la croissance a diminué. La tolérance à la salinité (à une eau de mer de 35,0 ‰) a été marginalement réduite dans le groupe de pH intermédiaires et considérablement réduite dans celui maintenu à bas pH, comparativement aux poissons témoins. L'activité de l'ATPase  $\text{Na}^+\text{K}^+$  a également été réduite dans les groupes de pH intermédiaires et bas. La régulation de  $\text{Na}^+$ ,  $\text{Cl}^-$  et  $\text{Ca}^{++}$  plasmatique a subi des effets adverses chez les groupes de pH intermédiaires et bas. Dans les conditions de notre expérience, la smoltification du saumon a été paralysée par des pH ambiants inférieurs à 5.





STUDIES ON ALUMINIUM TOXICITY IN THE PRESENCE OF HUMIC ACID

by

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1. 2. 3. 4. 5. 6. 7. 8. 9. 10.

$$\begin{aligned}
 & \text{1. } x^2 + y^2 = 1 \quad \text{2. } x^2 + y^2 = 1 \quad \text{3. } x^2 + y^2 = 1 \\
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 \end{aligned}$$

## ABSTRACT

Effect of humic acid on toxicity of aluminium in acidic waters was studied under laboratory conditions. ILL values at pH 5.0 were calculated to be 179 and 245  $\mu\text{g Al/L}$  with and without humic acid, respectively. The corresponding values at pH 5.5 were 188 and 169  $\mu\text{g Al/L}$ , respectively.

## RÉSUMÉ

L'effet de l'acide humique sur la toxicité de l'aluminium dans des eaux acides a été étudié dans des conditions de laboratoire. Les valeurs de ILL à un pH de 5,0 ont été calculées à 179 et 245  $\mu\text{g Al/L}$ , avec et sans acide humique, respectivement. Les valeurs correspondantes au pH de 5,5 ont été de 188 et 169  $\mu\text{g Al/L}$  respectivement.

Acidification of surface water is a major problem in parts of eastern Canada, especially in Nova Scotia. Runoff from acidic precipitation has added another dimension to the problem, since it may promote considerable mobilization of aluminium and trace metals like copper, zinc, cadmium, lead, etc., from the drainage basin to the lakes and streams (Cronan and Schofield 1979; Hermann and Baron 1980; Hall and Likens 1981). Several reports have been published indicating that aluminium mobilized by acidic water has contributed to fish mortality (Baker and Schofield 1980; Driscoll et al. 1980; Muniz and Leivestad 1980) in several parts of the world. Elevated levels of aluminium have also been reported in streams from Nova Scotia, and serious concern has often been expressed regarding the effect of aluminium on freshwater fishery resources. Most streams and lakes are also characterized by having waters of less than 5.4 pH and humic acid content in the range of up to 5 mg/L. The survival and growth of brook trout and white suckers during developmental periods, i.e. eggs, larvae, and postlarvae at pH levels 4.2 to 5.6, and aluminium concentrations up to 0.5 mg/L have been investigated (Baker and Schofield 1982). However, effects of natural organic chelators like humic acid have not been investigated. Consequently, a study was undertaken to determine the mitigating effect, if any, of humic acid, on toxicity of aluminium at several pH levels. Reduction in toxicity of metals like Cu by humic acid has been documented (Zitko et al. 1973).

Atlantic salmon parr ( $n = 10$ , avg. length = 8.2 cm) were used for 96-h bioassays in running tapwater in fiberglass tanks (32 L) at 10°C. Constant-head Mariotte bottles were used as required to deliver aluminium sulphate, sodium humate, and sulphuric acid and/or sodium hydroxide solutions to the test tanks. Concentrations of aluminium and humic acid were monitored several times a day throughout the tests. The tests were run at several pH's. Data for runs at pH 5 and 5.5 are given below:

Calculated lethal thresholds (ug/L) for Al and salmon parr

Toxicant	pH	
	5.0	5.5
Al	179	188
Al + sodium humate (5 mg/L)	245	169

The lethality lines (Fig. 1) indicate that, at lower concentrations of Al, the presence of humic acid does not affect the toxicity of Al but, above 1 mg Al/L, the toxicity is somewhat modified by it. At present, experiments are being conducted to determine toxicity of Al at 5°C and at several selected pH's. Field experiments, using water with natural humate, are also planned for future study.

Complexation of Al with humic acid in laboratory and field situations is also being investigated. At present, it is not known whether natural humates form complexes with Al as they do with other metals like Cu and Zn, with consequent reduction of toxicity to fish.

Three macroreticular resins (XAD-2, -4 and -7) of varying polarity have been tested for recovery of Al and humic acid from water, under laboratory conditions, and have performed satisfactorily. Nearly 100% recovery of Al is obtained with HCl,  $\text{NH}_4\text{OH}$  or NaOH. Humic acid is best retained by XAD-2 and can be eluted from the resin with alcoholic  $\text{NH}_4\text{OH}$ .

Al mixed with sodium humate loaded on XAD-2 and XAD-7 columns is separated into two fractions by an acid eluent followed by a base. The first fraction contains only Al while the second contains Al and humate, suggesting that a portion of Al may have formed a humate complex on mixing it with sodium humate. A number of humate samples have been collected from Nova Scotia lakes and the technique developed will be applied to determine whether Al is bound to humates under field conditions.

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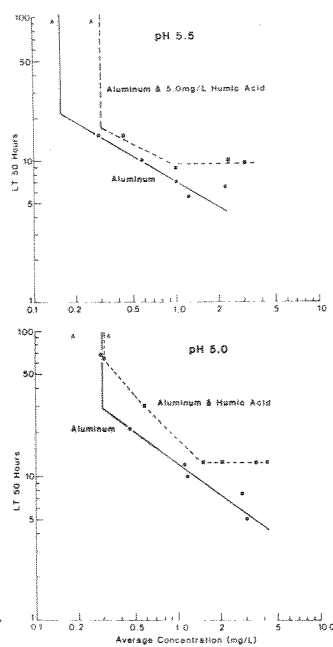


Fig. 1. Lethality line for juvenile salmon exposed to Al and to Al + humic acid at 10°C at pH 5.0 and 5.5.



ENERGY METABOLISM DURING SEVERAL LIFE STAGES OF  
SALMO SALAR UPON EXPOSURE TO LOW pH

by

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## ABSTRACT

Several studies are in progress to determine the effect of exposure to low pH on intermediary energy metabolism during several life stages of Atlantic salmon (Salmo Salar).

Fertilized eggs exposed to pH 4.5 through hatching to swim-up showed a larger cumulative mortality than those held at pH 6.5. Biochemical analyses are incomplete for this study.

Atlantic salmon parr were held at pH 4.5 during final stages of smoltification (83 d). The control salmon (pH 6.5) showed an increase in weight and length, and liver weight which were not observed in the low pH group. There was a larger decrease in condition factor of the low pH group than of the controls.

After 15 d, levels of ADP and glucose were higher and adenylate energy charge (AEC), creatine phosphate and glycogen were lower in the muscles of acid-exposed salmon. These qualitative differences were maintained until the end of the experiment. These results indicated that exposure to low pH caused gluconeogenesis from amino acid precursors through the citric acid cycle to become the predominant pathway rather than from glycogen.

The levels of ATP, total adenylates, AEC and glucose were consistently higher in the livers of the acid-exposed salmon compared to the controls. Together with the observed lack of growth, these results indicate that low pH caused impairment of anabolic processes.

## RÉSUMÉ

Plusieurs études sont en cours dont les objectifs sont de déterminer l'effet de l'exposition à de bas pH sur le métabolisme énergétique intermédiaire du saumon atlantique (Salmo salar) pendant plusieurs phases de son cycle de vie.

Les oeufs fécondés exposés à un pH de 4,5 depuis l'éclosion jusqu'au stade de nage vers le haut ont subi des mortalités cumulatives plus élevées que ceux maintenus à un pH de 6,5. Les analyses biochimiques ne sont pas encore terminées.

Des tacons ont été maintenus à un pH de 4,5 pendant les stades finals de la smoltification (83 jours). Les saumons témoins (pH 6,5) augmentèrent de poids et de longueur du corps et de poids du foie, augmentations qui n'ont pas été observées dans le groupe maintenu à bas pH. La diminution du coefficient de condition a été plus forte dans le groupe à bas pH que chez les témoins.

Après 15 jours, les niveaux d'ADP et de glucose ont été plus élevés, tandis que la charge énergétique d'adénylates (AEC), le phosphate de créatine et le glycogène ont été plus faibles dans les muscles de saumons exposés à l'acide. Ces différences qualitatives se sont maintenues jusqu'à la fin de l'expérience. Ces résultats indiquent qu'une exposition à un bas pH cause une gluconéogénèse à partir d'acides aminés comme précurseurs, passant par le cycle de l'acide citrique, pour devenir la voie prédominante plutôt qu'à partir du glycogène.

Les niveaux d'ATP, d'adénylates totaux, d'AEC et de glucose ont été uniformément plus élevés dans le foie de saumons exposés à l'acide, comparativement aux témoins. Ces résultats, joints au manque de croissance observé, indiquent qu'un bas pH nuit aux processus anaboliques.

## INTRODUCTION

Low pH of many freshwater systems is associated with acid precipitation or runoff from boggy areas. Acidification of lakes and streams is resulting in the disappearance of fish populations (Beamish and Harvey 1972; Schofield 1976). The sensitivity to toxic effects of low pH can vary with different life stages of *Salmo salar* (Daye and Garside 1977).

Adenylate energy charge (AEC) is a measure of the metabolic energy state of cells (Atkinson 1977). AEC is considered to be a potentially useful indicator of sublethal effects caused by xenobiotics or physiologically undesirable conditions (Haya and Waiwood 1983; Ivanovici 1980). Activities of regulatory enzymes of catabolic and anabolic pathways are altered in order to maintain a steady state value of AEC. For example, exposure to low pH causes a decrease in AEC of various tissues of *Fundulus grandis* (MacFarlane 1981) and alterations in other biochemical parameters of muscle energy metabolism in *Tilapia mossambica* (Murthy et al. 1981).

In the present study *Salmo salar* were exposed to low pH during two life stages, from fertilization of eggs to swim-up (alevin experiment) and during the parr-smolt transformation (smoltification experiment).

### ALEVIN EXPERIMENT

During previous work on the development of chorionase activity (Haya and Waiwood 1981; Waiwood and Haya 1983) a behavioral change in embryos held at low pH was observed. There was a definite lack of movement in these embryos which appeared to hinder their escape from the chorion ghost and a lack of movement once hatched. Peterson and Martin-Robichaud (1983) have shown that lack of movement in embryos about to hatch when held at low pH could be reversed on switching to ambient pH. The purpose of the current study is to determine AEC and related substrates of energy producing catabolic processes in Atlantic salmon alevins exposed to low pH during hatching until swim-up. The toxic effects of low pH on alevins may be related to disruption of intermediary energy metabolism.

### SMOLTIFICATION EXPERIMENT

Physiological and biochemical changes that occur during smoltification have been reviewed (Wedemeyer et al. 1980). The parr-smolt transformation does not proceed normally during exposure of salmon to low pH (Saunders et al. 1982). The purpose of this study was to determine the effects of low pH during smoltification on intermediary energy metabolism in order to obtain information on biochemical mechanisms of acid toxicity.

## MATERIALS AND METHODS

### ALEVIN EXPERIMENT

*Salmo salar* eggs were obtained from the North Atlantic Salmon Research Centre (NASRC). Eggs from four females were mixed with milt from six males and

were allowed to harden in Chamcook Lake water (all fish were grilse returning to NASRC). Within 4 h the eggs were transported to the Biological Station, St. Andrews, and placed in experimental containers. Approximately 10,000 eggs were divided between control (ambient pH) and low pH (4.5) boxes. For details of the experimental set-up see Waiwood and Haya (1983). Sampling was initiated at time of first successful hatching, i.e. 442 degree days. Alevins were grouped in the compartments according to age; since the numbers of alevins in low pH were much reduced by early mortalities, the groups at low pH span longer periods of time. A total of 1481 alevins at low pH were sampled and 3194 from control conditions.

Alevins to be sampled were removed from the experimental containers, quickly blotted dry and dropped into liquid nitrogen. The samples are stored at -80°C awaiting analysis for AEC.

### SMOLTIFICATION EXPERIMENT

The exposure of Atlantic salmon parr to low pH for our experiment was conducted as part of another project (Saunders et al., see this report). Thus a large number of salmon were exposed and our samples were chosen at random from the total pool of salmon in all treatment groups. Atlantic salmon parr were exposed to low pH or pH 6.5 (controls) as previously described (Saunders et al. 1982). The temperature was maintained at 10°C and the fish were fed commercial (EWOS) dry pellets at a rate of 2% body weight/day. The salmon were acclimated to the experimental conditions and apparatus. For the low pH group, the pH was decreased gradually over 11 d then maintained between 4.5 and 5.0 for the remainder of the experiment.

Periodically during the experiment (March 2, 1982-May 28, 1982) six salmon parr from each treatment were transferred with a dip net to 10 L of water of the same pH and temperature. The salmon were anaesthetized with t-amyl alcohol. The gills, liver and a section of white muscle (dorsal-lateral between the pectoral and anal fins) were rapidly excised, frozen with liquid nitrogen and stored at -80°C. The liver and muscle samples were analyzed, using enzymatic spectrophotometric techniques (Bergmeyer 1974; Haya et al. 1983) for adenosine triphosphate (ATP), adenosine diphosphate (ADP), adenosine monophosphate (AMP), creatine phosphate, glucose and glycogen. AEC was calculated from the formula:  $AEC = (ATP + 0.5 ADP) / (ATP + ADP + AMP)$ .

## RESULTS AND DISCUSSION

### ALEVIN EXPERIMENT

Cumulative mortalities during the experiment were 9.6% in the controls (pH 6.5) compared to 67.8% for the acid-exposed group (pH 4.5; Fig. 1). Numbers of dead included infertile eggs, embryos dead prior to hatching, those dying during the hatching process and alevins. A large number of deaths occurred during the early part of the exposure and was probably due to the inability of the eggs to adapt to pH 4.5. There was no difference between the degree days for the first successful hatching to occur in both treatments. The most sensitive period to the toxic effects of

low pH appeared to be during the hatching process (500-850 degree days).

Determination of the biochemical parameters of the alevins has not been accomplished. Sensitive analytical methods will have to be developed for the low levels of adenine nucleotides in these samples.

#### SMOLTIFICATION EXPERIMENT

A linear regression indicated a significant ( $p < 0.01$ ) increase in weight and length with time for the salmon held at pH 6.5 (controls) but not for salmon held at pH 4.5-5.0 (acid-exposed) over the 83 d of the experiment (Fig. 2). Saunders et al. (1982) reported a decrease in growth rate for salmon during smoltification at pH 4.5 compared to pH 6.5. The overall trend in our experiment was a decrease in weight of the acid-exposed salmon, thus a significant (linear regression,  $p < 0.01$ ) decrease in condition factor (CF) was observed (Fig. 2). A small decrease in CF is expected during smoltification (Love 1980; Saunders et al. 1982; Wedemeyer et al. 1980). The CF decreased significantly ( $p < 0.01$ ) from 1.05 to 0.92 in controls and to 0.83 in the acid-exposed group. The decrease of CF in the acid-exposed group was significantly more than that of the controls (ANOVA,  $p < 0.01$ ). This is in contrast to a similar experiment in which there was no difference in the CF between the two groups after smoltification was completed in the controls (Saunders et al. 1982). This discrepancy may be because the salmon parr in the Saunders' study were much larger at the beginning of the experiment compared to the parr of our study (mean of day 1 controls was 60.5 g vs 32.7 g; the small loss in weight would have a greater effect on the CF of smaller fish compared to larger fish). There was a significant (linear regression,  $p < 0.01$ ) increase in liver weight of the control salmon but not of the acid-exposed salmon. This resulted in a significant increase in the liver somatic index of the control and no significant change in that of the acid-exposed group (Fig. 2).

The decrease in CF of the controls may be due to an increase in the streamlining of the body during smoltification (Love 1980). However, the reduction of CF and altered metabolic energy pathways (see later) are indicative of a decrease in the physiological status of the acid-exposed group during our experiment.

The mean levels of adenine nucleotides (ATP=7.76; ADP=0.82; AMP=0.09;  $\mu\text{mol/g wet wt}$ ;  $n=6$ ) and AEC (0.937;  $n=6$ ) in the muscles of control salmon at the start of the experiment are similar to those reported for white muscle in other fish (Vetter and Hodson 1982; MacFarlane 1981; Jorgensen and Mustafa 1980). The mean level of ATP (1.07  $\mu\text{mol/g wet wt}$ ;  $n=6$ ) in the liver was lower than that found in muscles, but those of ADP and AMP were similar in the liver and muscle. Thus the mean AEC value was lower in the liver (0.689) and was comparable to that of killifish liver (MacFarlane 1981). The levels of adenine nucleotides in the gills could not be measured by spectrophotometric techniques and will require a more sensitive method of analysis.

A preliminary statistical evaluation of the biochemical data has been completed. Generally the biochemical parameters in the muscles (Fig. 3 and 4) and livers (Fig. 5 and 6) of control salmon varied

significantly but in a random fashion (ANOVA,  $p < 0.05$ ) with time and the variations were probably of no biochemical significance. However, there are some fluctuations that can be related to those weeks during which all the fish were subjected to several days of starvation and a day of anaesthesia and handling (sampling procedures for a concurrent experiment, see Saunders et al. this report). For instance, large decreases in liver glycogen occurred on days 41, 69 and 83 (Fig. 6). These days correspond to the sampling times for Saunders' experiment. Thus biochemical results from samples taken on these days were evaluated with caution. This may be the reason we were unable to detect the decrease in liver glycogen that occurs during the parr-smolt transformation (Wedemeyer et al. 1980).

Analysis of variance ( $p < 0.01$ ) indicated that there were differences between the levels of ATP, ADP, AEC, creatine phosphate, glucose and glycogen in muscles of acid-exposed salmon and those of controls. After 15 d of the experiment the levels of all of these except ATP differed in a consistent manner between the two groups. ADP and glucose levels were higher and AEC, creatine phosphate and glycogen levels were lower in muscles of the acid-exposed group compared to those of controls (Fig. 3 and 4). The large decrease of AEC observed in muscles of killifish (*Fundulus grandis*) upon acute exposure to low pH (MacFarlane 1981) was not seen in our experiment. In the muscle of *Tilapia mossambica* acute exposure to acid conditions decreased white muscle glycogen concentrations, but elevated glycogen levels were found after 15 d of exposure (Murthy et al. 1981). This is in contrast to our results in which the glycogen content remained lower for the 83 d of the experiment (except with day 48 samples, Fig. 4) in the muscles of acid-exposed salmon.

Two of the possible explanations for the decreased AEC and glycogen levels and the maintenance of fairly constant glucose levels in muscles of acid-exposed salmon compared to those in muscles of controls are: First, there could be an increase in rate of glycogenolysis to maintain glucose levels and a slight decrease in oxidative-phosphorylation resulting in a decrease of AEC. Second, there could be a decrease in glycogen synthesis and an increase in gluconeogenesis from amino acid (protein) precursors through the citric acid cycle rather than from glycogen. A simultaneous increase in glycolysis leads to a reduction in AEC because glycolysis (an anaerobic process) is less efficient than oxidative phosphorylation in the production of metabolic energy from glucose. The latter description of biochemical processes involved during the sublethal exposure of salmon to low pH is more likely because:

1. Exposure of fish to low pH has been suggested to cause hypoxia (MacFarlane 1981);
2. Hypoxia causes a reduction in AEC of fish muscle (Vetter and Hodson 1982) and of some worms (Schottler 1979) due to increased reliance on anaerobic metabolic pathways;
3. Gluconeogenesis with amino acids rather than carbohydrates as the initial substrate is the preferred route of maintaining glucose levels in fish (Walton and Cowey 1982).

AEC remained only slightly lower in the muscles of the acid-exposed salmon compared to those of the controls after 15 d of exposure to low pH (Fig. 3). AEC values in the acid-exposed group remained above 0.9 which is in the range expected for fish muscles in a healthy metabolic energy state (MacFarlane 1981; Vetter and Hodson 1982; Jorgensen and Mustafa 1980). Thus, there are sufficient metabolic energy stores to meet the requirements for mechanical work (muscle contraction) and osmotic work (active transport) and the observed biochemical effects in white muscle after exposure of salmon to low pH may not be of any physiological consequence.

The levels of ATP, total adenylates, AEC and glucose were consistently higher (ANOVA,  $p < 0.01$ ) in the livers of acid-exposed salmon than in those of control salmon. The metabolic energy of liver is used in active transport processes but is mainly required for anabolic processes. As in muscle, the high level of glucose in liver of acid-exposed salmon may be indicative of an increase in gluconeogenesis through amino acid rather than glycogen consuming pathways. However, the increased levels may also indicate a decrease in the ability to utilize glucose because of impairment of anabolism. Increased levels of ATP, total adenylates and AEC and no significant difference in creatine phosphate or glycogen stores between acid-exposed salmon and control salmon mean that a relative decrease in anabolism (energy consuming) compared to catabolism (energy producing) has occurred. This decrease in anabolic processes eventually results in a lack of growth and development. Thus, disruption of anabolism contributes to the detrimental effects caused by exposure of salmon to low pH during the parr-smolt transformation.

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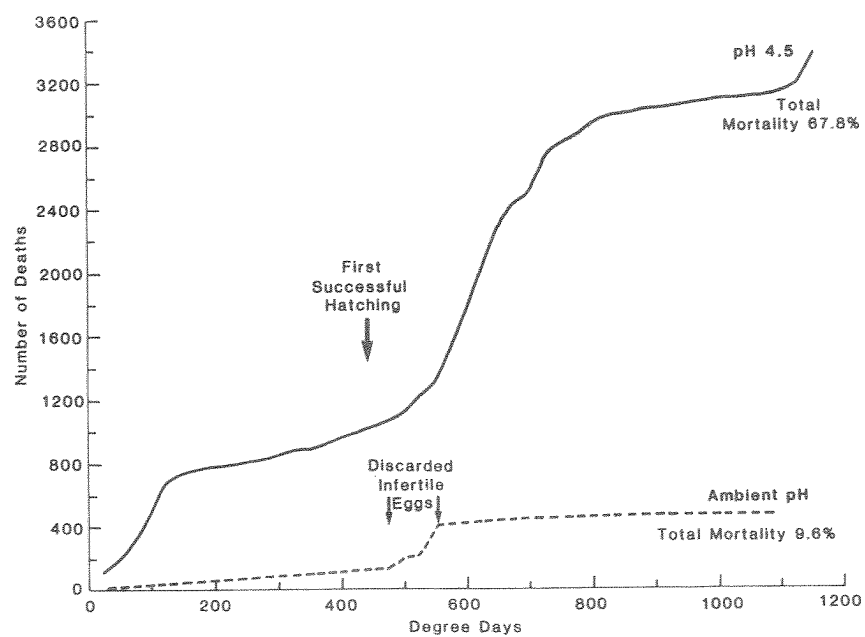


Fig. 1. Cumulative mortalities of Atlantic salmon from fertilization to swim-up at low pH (4.5) and ambient pH (6.5).

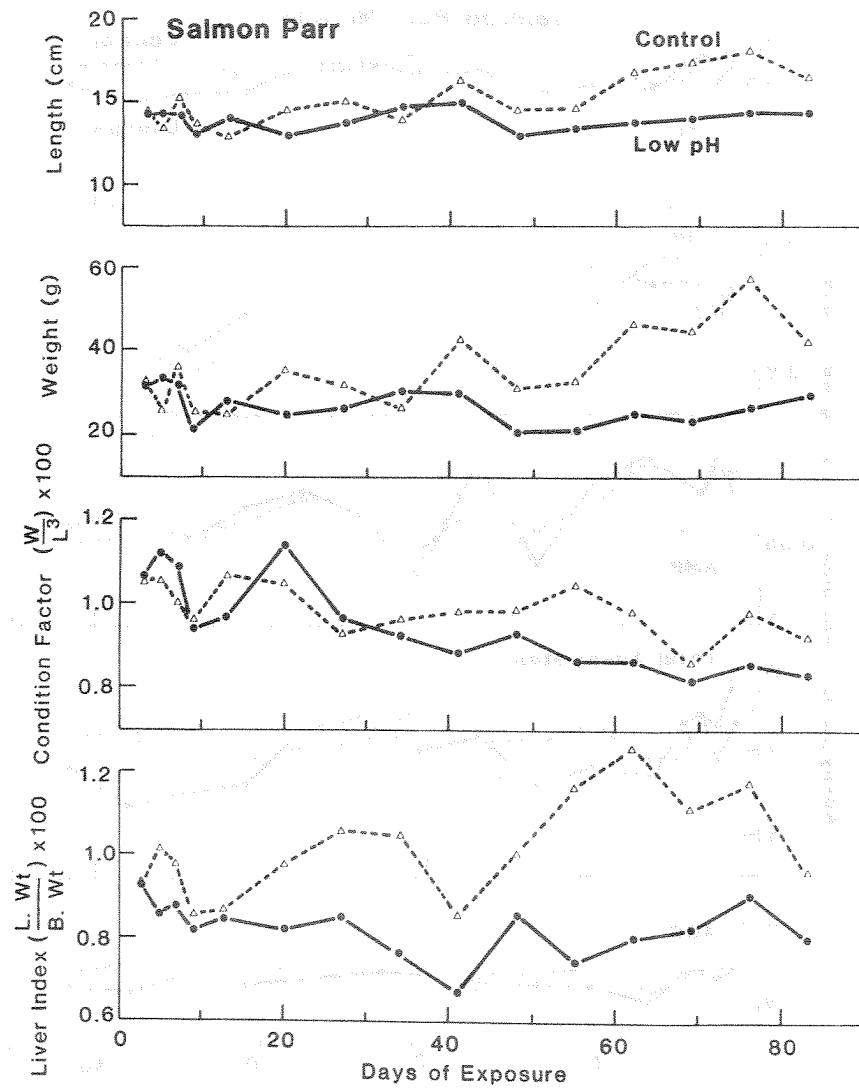


Fig. 2. Liver somatic index, condition factor, length and weight of Atlantic salmon during smoltification at low pH (4.5-5.0) and control pH (6.5).

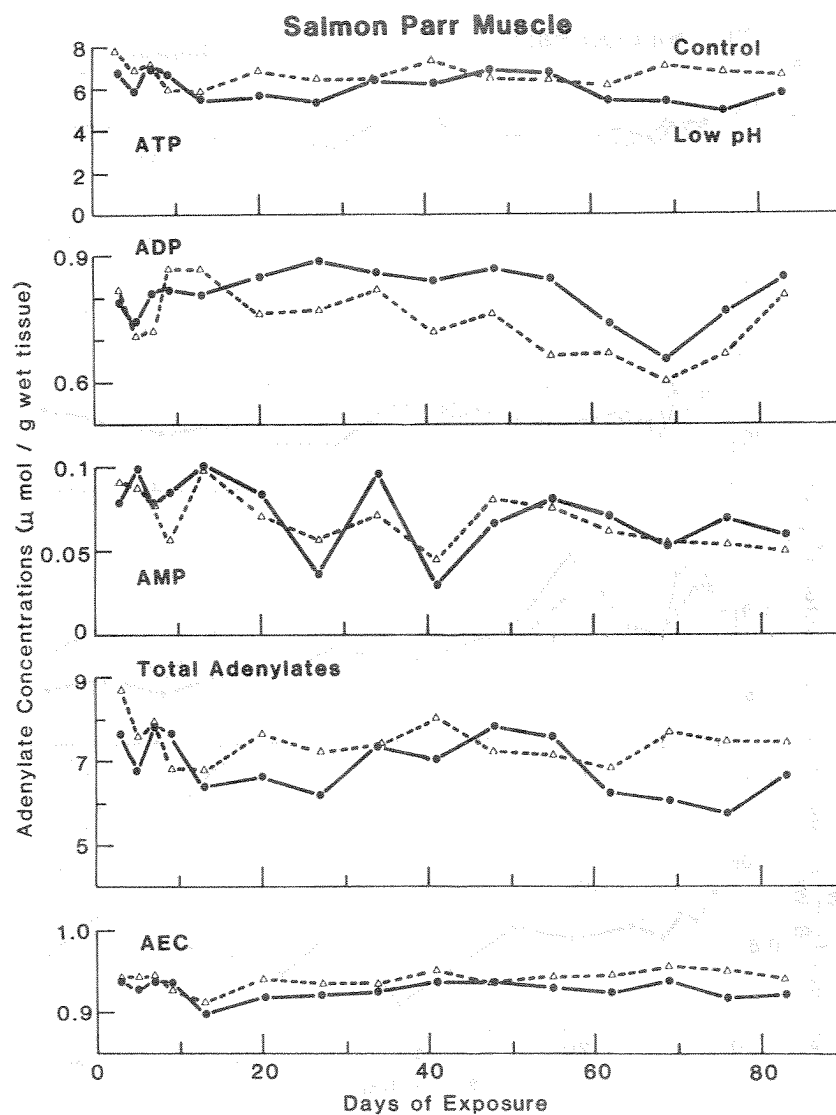


Fig. 3. Adenine nucleotide levels and AEC of Atlantic salmon muscle during smoltification at low pH (4.5-5.0) and control pH (6.5).



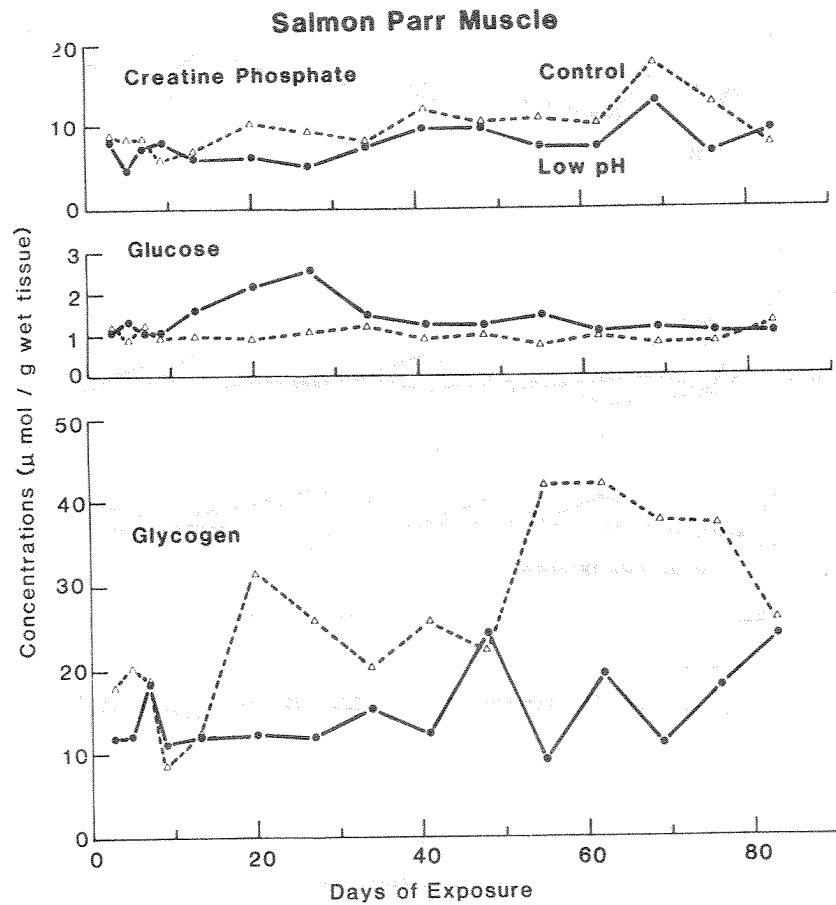


Fig. 4. Creatine phosphate, glucose and glycogen levels of Atlantic salmon muscle during smoltification at low pH (4.5-5.0) and control pH (6.5).

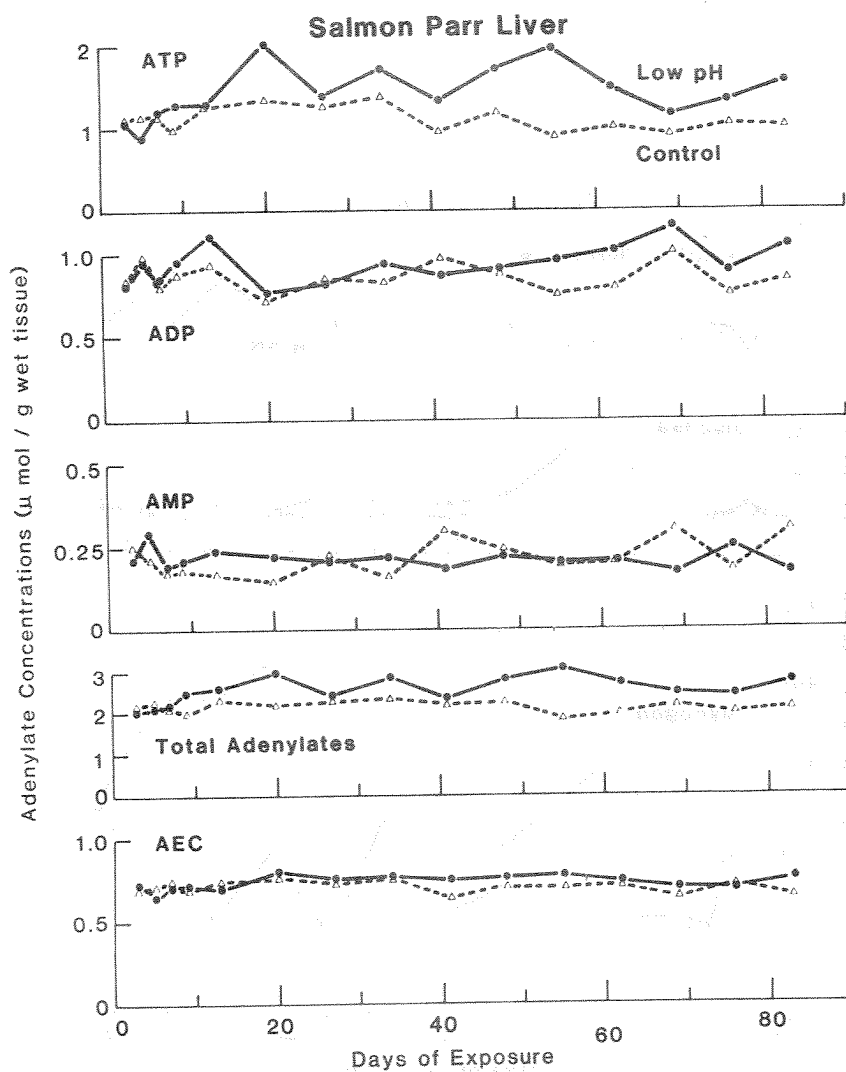


Fig. 5. Adenine nucleotide levels and AEC of Atlantic salmon liver during smoltification at low pH (4.5-5.0) and control pH (6.5).

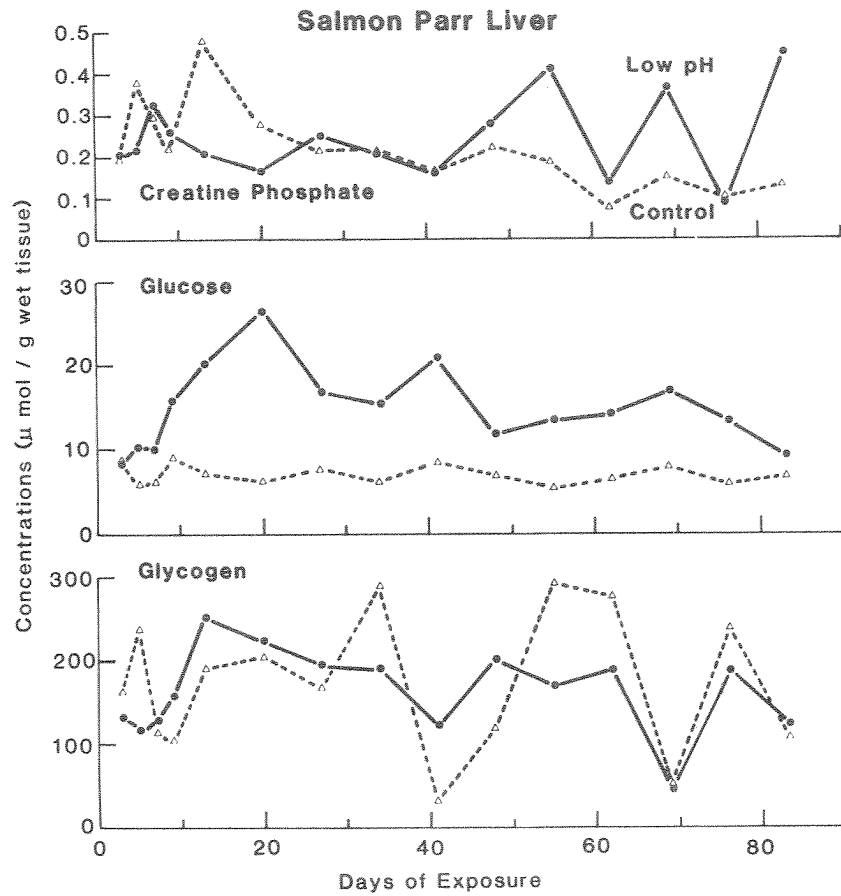


Fig. 6. Creatine phosphate, glucose and glycogen levels of Atlantic salmon liver during smoltification at low pH (4.5-5.0) and control pH (6.5).



EFFECTS OF ACIDIC pH ON THE GROWTH AND BEHAVIOR OF BLACKNOSE DACE, SLIMY  
SCULPINS AND JUVENILE ATLANTIC SALMON IN A SIMULATED STREAM ENVIRONMENT

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## ABSTRACT

Preliminary results (after the completion of three of sixteen replicates) are presented for an investigation of effects of sublethal levels of acidic pH (5.0-6.5) on growth and intra- and interspecific behavior of the blacknose dace, slimy sculpins and juvenile salmon. Tests were conducted in a simulated stream environment at two stream velocity regimes. At all treatment levels, interspecific interactions have been similar; juvenile salmon usually hold stations on the stream bottom and are territorial and the dace and sculpins consistently flee when attacked by salmon. No predation of any species has been observed. A positive relationship between weight gain and pH is indicated for all species. No differences in feeding frequency at the different treatment levels were observed for wild salmon parr and hatchery reared fry; however feeding frequency of wild fry appears to decrease with declining pH.

## RÉSUMÉ

Nous présentons ici les résultats préliminaires (après achèvement de trois sur seize répliques) d'une étude sur les effets de niveaux sublétaux de pH acides (5,0-6,5) sur la croissance et le comportement intra et interspécifique de naseux noirs, de chabots visqueux et de saumons juvéniles. Les essais ont été menés dans un environnement simulant un cours d'eau, à deux régimes de vitesse de courant. Les interactions interspécifiques sont identiques, à tous les niveaux de traitement; les saumoneaux maintiennent ordinairement leur station sur le fond et exhibent un comportement territorial, tandis qu'invariablement les naseux noirs et les chabots visqueux fuient lorsque attaqués par les saumons. Nous n'avons observé de prédation chez aucune espèce. Il y a, chez toutes les espèces, une relation positive entre le gain pondéral et le pH. Nous n'avons pas observé de différence de fréquence de l'alimentation aux différents niveaux de traitement entre des tacons sauvages et des alevins de saumon élevés en pisciculture, bien que cette fréquence, chez des alevins sauvages, semble diminuer à mesure que baisse le pH.

## INTRODUCTION

On the basis of toxicological investigations, Daye and Garside (1980), and Daye (1980) have suggested that Atlantic salmon should not be stocked in waters having a pH of less than 5.0. Although there is conflicting evidence as to the effects of pH on fish growth (Fromm 1980), pH levels above 5.0 may retard the growth of juvenile salmon (Farmer et al. 1980; Spry et al. 1981). The aim of this study is to investigate the effects of sublethal levels of acidic pH on the behavior and growth of juvenile salmon (wild and hatchery-reared fry and wild parr), and two sympatric species, blacknose dace (*Rhinichthys atratulus*), and slimy sculpins (*Cottus cognatus*) that are potential competitors with salmon for food and space. Interspecific as well as intra-specific interactions are being considered to assess whether changes in competitive relationships among the species occur at different pH levels.

## MATERIALS AND METHODS

The experiments are being conducted in four large stream tanks in which natural substrate, light, temperature, stream flow and fish population density conditions are simulated. For each experiment, each tank is maintained at the same current velocity regime but a different pH level - one of the four treatment levels, i.e., a pH of 5.0, 5.5, 6.0 or 6.5. The pH effects are being assessed at two velocity regimes, a "high" (1-60 cm/sec) and a "low" (1-30 cm/sec) regime, used alternately in the experiments. Water temperature, pH, and heavy metal levels are regularly monitored. The fish in each tank are fed, twice daily, a standard ration of brine shrimp and tubifex worms.

At the start of each experiment, all fish are anesthetized with tertiary amyl alcohol, and then weighed, measured (fork length), and individually branded (as per the fin-branding method of McNicol and Noakes (1979)).

The following is the experimental protocol used in each experimental replicate: For the first week, only wild salmon parr and fry and non-salmonid fishes are maintained in the stream tanks. On the first, second, fourth and seventh day of this week, observations on each fish are made once in the morning and again in the afternoon. Each fish is located and then its behavior for the next minute is noted. In order to simulate the situation where hatchery-reared fish are stocked in a stream, they are placed in the test facility after the other fish have been allowed to establish prior residence. Hatchery-reared salmon fry are introduced on the morning of the eighth day (the start of the second week). On each morning and afternoon of 10 test days following the introduction of the hatchery-reared fry, the location and behavior of all fish (for a 1-min period) are again recorded. Additionally, on each test day more lengthy observations are made of one fry (15 min) and the single parr (6 min) in each tank. The identity of the one fry (of the four in each tank) to be observed in the 15-min observation is predetermined in a randomized manner prior to the start of each replicate of the experiment. After the observations for each replicate have been completed, the tanks are emptied, and the fish are collected and reweighed and measured.

## RESULTS

At all pH levels to date, interspecific interactions have been similar. Fry, particularly the wild fish, actively defend areas of the tank and attack other fry, blacknose dace, sculpins and, occasionally, parr that linger near them. Blacknose dace and sculpins consistently flee when attacked and sometimes are pursued for short distances. Chases typically end when the fish have left the area (dace) or have hidden among rocks on the bottom (sculpins). No social interactions between dace and sculpins have been observed although the two species have sometimes been seen close together in the tanks. The parr, although maintaining stations, less consistently defend the area around them. Sometimes fry, dace and sculpins are tolerated at close range; at other times they are attacked and driven out of the defended area. No predation of either fry or sculpins has occurred; however three of the 32 dace used in the first two replicates of the experiment were not found. These may have been eaten but no extended pursuit of the dace by the other fishes has been observed.

Six of the twelve sculpins tested at the lowest pH (5.0) died, but few other deaths (for any species) have occurred. Preliminary results (after three of 16 replicate experiments have been completed) suggest a positive relationship between weight gain and pH for all species. Feeding frequency was enumerated only for the Atlantic salmon fry and parr. No differences in feeding frequencies among treatment levels were observed for the parr and the hatchery fry; however, for wild fry, feeding frequency appears to decrease with declining pH. All fish except the sculpins readily capture the food supplied; the sculpins are less adept at handling drifting material (i.e. when swimming after food they miss it more frequently than do the other species).

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EMBRYO MOVEMENTS OF ATLANTIC SALMON AS INFLUENCED BY pH,  
TEMPERATURE, AND STATE OF DEVELOPMENT<sup>1</sup>

by

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<sup>1</sup>A complete account of this work is to be published elsewhere.



## ABSTRACT

Two types of embryo movements of Atlantic salmon (*Salmo salar* L.), pectoral fin flutter and trunk movements, were measured as a function of incubation temperature, pH, and developmental state. Trunk movements began at 200 degree days, initially at highest frequency. The frequency decreased throughout subsequent development, then increased slightly just before hatching. Movements were much more stereotyped in the earliest stages of development than just prior to hatching. Frequencies at 200 degree days were insensitive to temperature and pH. Frequencies just prior to hatching were very temperature sensitive ( $Q_{10}$  of 13) and were decreased at low pH. This decreased frequency may be related to documented effects of temperature and low pH on hatching. Pectoral fin movements were temperature sensitive ( $Q_{10}$  of 2) and pH insensitive. These movements were initiated at 350 degree days of development and attained maximal frequency at 400 degree days.

## RÉSUMÉ

Deux types de mouvements d'embryons de saumon atlantique (*Salmo salar* L.), les battements des nageoires pectorales et les mouvements du tronc, ont été mesurés en fonction de la température d'incubation, du pH et de l'état de développement. Les mouvements du tronc commencent à 200 jours-degrés, la fréquence étant maximale au début. Elle diminue par la suite, pour augmenter légèrement juste avant l'éclosion. Les mouvements sont plus stéréotypés dans les premiers stades de développement que juste avant l'éclosion. La fréquence, à 200 jours-degrés, n'est pas influencée par la température et le pH. Elle est par contre très sensible à la température ( $Q_{10}$  de 13) juste avant l'éclosion et diminue quand le pH est bas. Il peut y avoir corrélation entre cette diminution de la fréquence et l'effet documenté de la température et du bas pH sur l'éclosion. Les mouvements des nageoires pectorales sont sensibles à la température ( $Q_{10}$  de 2), mais insensibles au pH. Ces mouvements commencent à 350 jours-degrés de développement et atteignent une fréquence maximale à 400 jours-degrés.