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# A Proposal for Environmental Research and Monitoring of Organic Pollution Caused by Salmonid Mariculture in the Bay of Fundy

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Biological Station St. Andrews, N. B. E0G 2X0

March 1990

Canadian Technical Report of Fisheries and Aquatic Sciences No. 1724

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A PROPOSAL FOR ENVIRONMENTAL RESEARCH AND MONITORING
OF ORGANIC POLLUTION CAUSED BY SALMONID MARICULTURE IN THE BAY OF FUNDY

by

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This is the two hundred and sixth Technical Report of the Biological Station, St. Andrews, N. B.

© Minister of Supply and Services Canada 1990 Cat. No. Fs 97-6/1724 ISSN 0706-6457

Correct citation for this publication:

Wildish, D. J., J. L. Martin, R. W. Trites, and A. M. Saulnier. 1990. A proposal for environmental research and monitoring of organic pollution caused by salmonid mariculture in the Bay of Fundy. Can. Tech. Rep. Fish. Aquat. Sci. 1724: iii + 24 p.

#### **ABSTRACT**

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Presented is our concept of the environmental problems likely to be encountered as the culture of salmonids in seawater net-pens becomes more intensive in the Bay of Fundy. Omitted from consideration are disease transmission and contaminant chemicals (such as therapeutants inclusive of antibiotics, pesticides, hormones and antifoulants used in construction materials). This proposal is concerned with the environmental results caused by the presence of high densities of salmonids in cages. Organic wastes which originate there may cause site-specific effects which result in buildup of a "mariculture sludge" near, and under, net-pen sites. Alternatively, it may cause a more general increase in the concentration of plant nutrients, such as ammonia, nitrate and phosphate, in seawater which may stimulate localized microalgal blooms. Research and monitoring methods are suggested which should allow the environmental effects anticipated to be temporally assessed as well as contributing ultimately to a predictive management model suitable for determining the holding capacity of salmonid culture areas in the Bay of Fundy.

### RÉSUMÉ

Wildish, D. J., J. L. Martin, R. W. Trites, and A. M. Saulnier. 1990. A proposal for environmental research and monitoring of organic pollution caused by salmonid mariculture in the Bay of Fundy. Can. Tech. Rep. Fish. Aquat. Sci. 1724: iii + 24 p.

Le rapport présente notre concept des problèmes environnementaux qui surviendront probablement à mesure que s'intensifiera la culture des salmonidés dans des parcs marins en filet, dans la baie de Fundy. Nous n'avons pas considéré les problèmes de transmission des maladies et des produits chimiques contaminants comme les produits thérapeutiques, y compris les antibiotiques, les pesticides, les hormones et les produits antisalissure dont on se sert dans les matériaux de construction. La présente proposition s'intéresse aux conséquences pour l'environnement de la présence de fortes densités de salmonidés dans les cages. Les déchets organiques qui en découlent peuvent avoir une incidence particulière sur les sites ayant pour effet d'entraîner une accumulation de bouillie de mariculture à proximité des emplacements des parcs ainsi qu'en dessous de ceux-ci. Cela pourrait aussi entraîner une augmentation plus généralisée de la concentration, dens l'eau de mer, d'éléments nutritifs pour les plantes comme l'ammoniaque, le nitrate et le phosphate, ce qui pourrait en retour stimuler la prolifération localisée de poussées d'algues microscopiques. Nous proposons que l'on procède à des recherches et que l'on instaure des méthodes de contrôle pour faire une évaluation temporelle des effets environnementaux anticipés et finalement pour prédire un modèle de gestion capable de déterminer la capacité des secteurs de culture des salmonidés dans la baie de Fundy.

#### INTRODUCTION

Within the Bay of Fundy, the growout of salmonids in seawater net-pens is a relatively recent and successful economic venture. mariculture industry is concentrated in the Western Isles region of the mouth of the Bay of Fundy in Charlotte County, New Brunswick, Canada. In 1988, this industry was still expanding due to increases in production at already established sites. During 1989, new sites were occupied consequent on the lifting of a moratorium on site applications imposed by the Provincial Department of Fisheries and Aquaculture. In the future it is possible that more seaworthy cages will be deployed in the offshore regions of the Bay, as well as further expansion in inshore regions. increased production levels implied by this expansion entail increased environmental stress, we consider it important to bring to the attention of the industry the likely environmental problems it will face in the near future to stimulate thought regarding a management strategy to minimize them.

Growout of salmonids in the sea is an intensive form of monoculture with biomass reaching 15-20 kg wet fish m3 of seawater within the net-pens. High densities of salmon growing at the maximum rate produce large quantities of wastes. The wastes include dissolved and particulate components consisting of faeces, ammonia excreted from the gills and uneaten food. The chemical composition and amounts of these substances depend on the type and amount of food supplied to the fish. Both the food and excretory wastes contain nitrogen and phosphorous compounds which are plant nutrients in the dissolved state and thus agents of eutrophication (see Ketchum 1969). Recently, in British Columbia, mass mortalities of cultured salmon were attributed to microalgal blooms of Chaetoceros convolutus Heterosigma akashiwo (Anon. 1988). southern Norway during 1988, a bloom of the diatom Chrysochromulina polyepsis caused mortality of cultured salmon as well as many other wild finfish (Granmo et al. 1988). A red tide bloom of Alexandrium excavata also caused direct mortalities of cultured salmon in the Faroe Islands (Mortensen 1985). In all these cases, nutrient enrichment derived from the breakdown of organic wastes from the salmon mariculture industry was considered to be one of a number of other factors contributing to the bloom. Typical nitrogen budgets for an actively growing

salmonid (Hakanson et al. 1988) show that 25% is retained by the fish during growth, 15% is lost as particulate faeces, and 60% leaves the gills as ammonium compounds dissolved in seawater. Thus, 75% of the nitrogen introduced as fish food is readily available for plant growth stimulation. In addition, a variable proportion of food fed (from 1-30%) remains unconsumed during feeding (Rosenthal et al. 1988).

Particulate food and faecal wastes may reach net depositional sediments beneath or near net-pens. Where erosional processes are low or absent, a buildup of wastes may occur and result in a biological oxygen demand which exceeds the sedimentary supply of oxygen. In such anoxic sediments, the microbial community is anaerobic with a respiration that first involves some form of nitrate reduction producing reduced nitrogen products. As the organic input is increased, the microbial community develops into one in which sulphate reduction and production of hydrogen sulphide dominates, followed finally by a microbial community where process the dominant respiratory methanogenisis and the chief product is The typical products of anaerobic methane. respiration from anoxic sediments are gases and usually all three types of respiration are at each stage of development, thus giving anoxic sediments the characteristic smell of rotten eggs due to the presence of hydrogen sulphide gas.

Environmental problems associated with salmon mariculture in other parts of the world have been described, e.g. in Scotland (Gowen and Bradbury 1987; Anon. 1988) and the Nordic countries (Ackefors and Södergren Hakanson et al. 1988). An International Council for the Exploration of the Sea Working Group on the environmental impact of mariculture has produced reports (Rosenthal et al. 1988; Anon. 1989). General areas of concern in these organic pollution, reports were chemical contaminants used by the industry inclusive of therapeutants, pesticides, hormones, antifoulants used in construction materials, and disease outbreak and transmission.

The purpose here is to present an environmental research and monitoring plan designed to assist in managing the salmonid mariculture industry in the Bay of Fundy. Only organic pollution is considered in this work and environmental problems posed by chemical contaminants and disease transmission are

excluded from further consideration. This presentation gives details of the sampling and analytical methods used in all environmental monitoring considered to be important for environmental management purposes and suggests a few research topics which must be addressed before a realistic management model for the Bay of Fundy salmon mariculture industry can be implemented.

## ENVIRONMENTAL RESEARCH AND MONITORING: A STRATEGIC PLAN

A fundamental and useful concept in ecology is that of "carrying capacity": the population size or steady state above which a limiting factor inhibits further population increase It is usually applied to (Gilpin 1976). populations in which the trophic resource is an integral part of the environment, as with a bed of blue mussels grazing the natural seston in a bay. Where the food is introduced by man, and never itself limiting, as in salmonid mariculture, it is considered useful to distinguish the situation by referring to the "holding capacity" of a salmon farm in a defined area of the marine ecosystem. The holding capacity of an area, such as Lime Kiln Bay, is simply the maximum population size which can be grown out there before inhibiting environmental factors, such as reduced dissolved oxygen, limit the population. of that production considerations involved in this concept include the extremes in the natural environment, inclusive of seasonal variance in numbers and biomass of commercially valuable biological resources, e.g. herring, it can be appreciated that the input data required is quite extensive.

We consider holding capacity models for defined sites or oceanographically defined areas of the Bay of Fundy salmonid culture industry to be practical resource management tools that can be used to determine the optimum numbers, or production, that are possible. models require multidisciplinary inputs of reliable physical, chemical and biological data, inclusive subcomponents. pelagic and benthic Ecosystem models of this type have previously been devised (see Gordon et al. 1986, 1987) and it should be possible to adapt them to model the flux of, say, dissolved oxygen, which is of great importance for the optimum growth of salmon. Modelling plant nutrients may also be possible and, because an increase may cause a microalgal bloom which, even if it is not itself toxic, will, by its death and decomposition, result in a localized area of dissolved oxygen depletion (Ketchum 1969).

Because model predictions always need to verified, we recommend a concurrent environmental monitoring project. The primary purpose of this would be to provide a temporal record of environmental conditions within the Letang-Western Isles area as a basis on which to make management decisions. The variables to be monitored include temperature, salinity, dissolved oxygen, plant nutrients, microalgal species and their abundances, and sedimentary conditions, inclusive of percent water, silt-clay and volatile solids content. Recommended methods of sampling and analysis will be found in the following sections.

# ANNUAL ENVIRONMENTAL SALMON NET-PEN SITE ASSESSMENTS

The purpose of measuring environmental variables at mariculture sites is to help make it possible to operate at, or near, the optimum holding capacity. To do this, it is necessary to determine some key-rate variables which are needed as input in a holding-capacity model. Every effort has been made to minimize the work involved so that data of little use are not collected. We provisionally recommend that the following be obtained annually for each mariculture operation: site location, site depth, annual production data and some environmental parameters. Site evaluations should apply to planned and existing operations and be annually updated in late summer. It is considered to be very important to make these observations and measurements in a standard way as follows:

<u>Site location</u> - accurately draw the position of existing cage structures on the standard chart of the area supplied by the Provincial Department of Fisheries and Aquaculture.

<u>Site depth</u> - measured near low water with a lead line marked off in units of 0.5 m. Record the depth of water to the nearest 0.5 m at a recorded date and time. Minimum of 5 and maximum of 50 depth/times taken from the cage site walkways. Record the position of each observation on the chart provided.

<u>Production data</u> - because there will usually be two year-classes present: smolts and market fish, the number and wet weights (estimated from the average weight of a subsample) in tonnes of each at the beginning of the growing

season and at its end or at the time of marketing. The weights of food, wet and dry separately, with food formulation used indicated, will be required for the same period. Average growth rates and percentage of mortalities of each year-class should also be available on an annual basis.

Environmental measurements measurements should be made during calm wind conditions near low-water slack during the (preferably August/September) taking surface seawater samples (minimum of 5 and up to 25) from the cage-site walkways. For each sample the temperature, salinity, and dissolved oxygen concentration are determined. Five to 25 sediment samples are taken and a subsample retained in a plastic whirl pack bag. Record the position of seawater and sediment samples on the chart. Record the general current direction and speed of surface seawater by use of a floating object and by timing it over a known distance.

#### SEAWATER SAMPLING

Take samples from the surface water in a clean, plastic bucket within, or just outside, a net-pen. The temperature of the seawater is determined immediately with a thermometer readable to 0.1 C and subsample of 100-200 mL removed for determination of salinity by argentometric titration or conductimetric method (the Autosal used at the Biological Station utilizes a conductimetric method which is reliable and semiautomatic, but expensive to purchase initially). A separate seawater sample is taken from the bucket for determination by Winkler titration of the dissolved oxygen content. Special BOD bottles are rinsed with the seawater before filling by immersion to exclude air bubbles.

#### SEDIMENT SAMPLING

Sediments must be collected quantitatively with a SCUBA-held corer (Plexiglass tube of 5-10 cm diameter which can be plugged before retrieval with a suitable cap or bung) or a grab operated from a winch on the sampling vessel. Care must be taken with all samples to discard those which do not maintain the surface sediment layer intact. Sampling is repeated until a satisfactory sample is obtained and a subsample removed from the top ca. 10 cm of the profile. The wet sample of up to 200 g (but only a few grams are needed if the silt-clay

content is excluded) is loaded into a well labelled, plastic, whirl pack bag or small glass bottle and temporarily stored in a cooler chest containing ice. For longer-term storage, the samples may be kept indefinitely in a freezer.

#### LABORATORY ANALYSIS

#### **Temperature**

A mercury thermometer readable to 0.1 C is used in the field. It is regularly checked against two independent thermometers to minimize the chance of systematic error, and calibrated in ice water.

#### Salinity

By low precision, argentometric titration according to Strickland and Parsons (1968). The following is required:

Glass sample bottles with tight-fitting screw cap.

International Standard Sea Water from Depot d'Eau Normale, Laboratoire Hydrographique, Charlottenlund Slot, Copenhagen, Denmark. Secondary standards may also be prepared by bottling filtered (Whatman GF/C filters) seawater stabilized by adding a few crystals of thymol.

25-mL brown glass, burette which is automatically zero-adjusting. Conical flasks with wide mouth and 200-mL capacity.

Magnetic stirring system.

Silver nitrate solution - dissolve 49 g of Anlar grade silver nitrate in 1 L of distilled water.

Indicator solution - dissolve 3.5 g of Anlar  $K_2CrO_4$  in 1 L distilled water.

Automatic pipets - 10 mL and 15 mL.

Add 10 mL of sample by automatic pipet and 15 mL of indicator solution to the flask. Titrate this solution with silver nitrate in the burette until the greenish color becomes a full yellow and pale red as the end-point is exceeded. Record the burette reading to 0.01 mL. As long as all reactants are kept at laboratory temperature for 24 h prior to analysis, there should be little

need for temperature correction. We also do not recommend using the calibration procedure given by Strickland and Parsons (1968) for silver nitrate solution. The primary standard has a chlorinity of exactly known value so the volume of silver nitrate:

(mL AgNO<sub>3</sub>) standard ≡ Cl o/oo

can be used to determine the unknown (Tait 1968):

(mL AgNO<sub>3</sub>) unknown  $\equiv$  Cl o/oo by:

Y CI o/oo = CI o/oo  $\frac{\text{mL AgNO}_3 \text{ unknown}}{\text{mL AgNO}_3 \text{ standard}}$ 

To convert from chlorinity to salinity (S o/oo):

 $S \circ 0/00 = 0.030 + (1.805 \times Cl \circ 0/00).$ 

Although it is widely used, this Knudsen equation does not have universal applicability and may even give misleading equivalents if applied to diluted seawater other than that used by the Laboratoire Hydrographique, Copenhagen.

# Dissolved oxygen by modified Winkler method (Strickland and Parsons 1968)

The dissolved oxygen concentration of seawater represents a dynamic state affected by many physical and biological processes which occur in natural seawater (see Fig. 1). It is important, therefore, to stabilize the sample as soon as possible after it has been taken. The following is required:

300-mL clear glass BOD bottles with ground-glass stoppers and etched numbers for identification

Burette - we use a Radiometer Autoburet Model ABUII with a digital readout to 0.01 mL

50 mL pipets

125 mL conical flasks

Manganous sulphate solution. Dissolve 48.0 g Anlar MnSO<sub>4</sub>.4H<sub>2</sub>O in 1 L distilled water

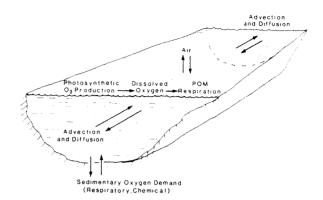


Fig. 1. Three-dimensional diagram of the dissolved oxygen balance in part of the Letang.

Alkaline iodide solution. Prepared by dissolving 500 g NaOH in 0.5 L distilled water and 300 g KI in 0.450 L distilled water and mixing with vigorous stirring until clear. Both reactants should be of analytical quality.

The above two reactants are dispensed from plastic, dropping bottles supplied with a plastic pipet marked at 1 mL and adjusted by means of a rubber bulb

Concentrated sulphuric acid of specific gravity 1.84

Approximately 0.01 N sodium thiosulphate solution Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub>.5H<sub>2</sub>O Anlar grade.

Add 2.0 g per L distilled water with 0.1 g Na<sub>2</sub>CO<sub>3</sub> and 1 drop of carbon disulphide as a preservative.

Starch indicator. Prepared by suspending 2 g soluble starch in 300 mL distilled water. Add a 20% solution of NaOH with vigorous stirring until clear. After 2 h, add concentrated HCl until the solution is just acid to litmus paper and then add 2 mL glacial acetic acid. Add distilled water to make 1 L. Discard this stock solution if it gives a greenish or brownish rather than a blue color near the end-point.

In the field and immediately after taking the bubble-free seawater sample, 1 mL of manganous sulphate and 1 mL of alkaline iodide are added and thoroughly shaken after restoppering the BOD bottle. On return to the

lab, and as soon as possible after taking the samples (always <24 h), but after allowing them to come to the laboratory temperature, 1 mL of concentrated sulphuric acid is added. sample bottle is restoppered and mixed by shaking until all the precipitate dissolves. 50 mL of the sample is transferred into the conical flask within 1 h of acidification and titrated with 0.01N thiosulphate until a pale straw color remains. 5 mL of starch indicator solution is added and the titration continued until the solution is colorless. The titration volume (V) for each sample batch is corrected by a blank titration volume (v), determined as above except that filtered seawater is used through which nitrogen gas has been bubbled to remove all of the oxygen (Parsons et al. 1984).

The corrected volume, V, is given by (V-v) mL and the dissolved oxygen content is determined as:

mg-atoms 
$$O_2/L = 0.1006 \times f \times V$$
 ........1

where the constant in equation 1 refers only to a 50-mL sample and a 300-mL BOD bottle. The factor f in equation 1 represents a calibration of the thiosulphate solution and is characteristic of each batch. It is determined by making a 0.01N potassium iodate solution by adding 0.3567 g dry KIO3 (dried at 105 C for 1 h) to 200 mL distilled water with warming. After the solution has cooled, add distilled water to 1 L. Fill a BOD bottle with distilled water, add 1 mL concentrated  $H_2SO_4$ , 1 mL alkaline iodide and mix thoroughly. Add 1 mL manganous sulphate solution and mix. Place 50-mL aliquots in conical flasks and add 5 mL 0.01N KIO3 with 0.01N thiosulphate stock solution. Theň

$$f = \frac{5.00}{\text{mL of thiosulphate added}}$$

The dissolved oxygen values determined for unknowns can be expressed as follows:

mg 
$$O_2/L = 16 \text{ x mg-atoms } O_2/L \dots 2$$

mL/L or mL/kg 
$$O_2$$
 at STP = 11.20 x mg-atoms  $O_2$ /L .....3

From an equation given by Weiss (1970), it is possible to calculate the expected concentration,

C\*, in mL/L of dissolved oxygen at a pressure of 1 atmosphere when the salinity (S o/oo) and absolute temperature, T (to convert °C to absolute temperature, add 273.15) are known. Thus:

Weiss (1970) also provides a table showing equilibrium dissolved oxygen values in mL/L at 1 atmosphere pressure and at different temperatures and salinities. Using the two latter parameters, it is possible to interpolate C\* from this table (included here as Appendix I as this may be more convenient to use). The percentage saturation at sea-level pressure is then:

Percent dissolved oxygen saturation =  $\frac{\text{mL/L of unknown}}{\text{C}^*}$  100 .....5

Frequent standardization of the technique is vitally necessary and this should be done with filtered seawater samples of known dissolved oxygen content to guard against systematic errors. A dissolved oxygen meter, such as the YSI Model 65, is useful in preparing seawater of approximately known O2 content, e.g. 0 and 50% oxygen content by passing nitrogen gas through the sample and 100% or oxygen saturated seawater by passing air bubbles through temperature controlled seawater. It should be remembered that the dissolved oxygen meter itself is prone to systematic errors, e.g. probe poisoning with sulphides, physical damage to the membrane, and operator bias due to inadequate calibration.

#### Sediment analysis

Three rapid analyses are suggested here which yield practical measures of sedimentary conditions. For a more complete analysis of sediments, see Holme and McIntyre (1984).

The equipment required (Akagi and Wildish 1975) includes the following:

Analytical balance readable to 0.1 mg
Oven capable of heating to 105°C
Muffle furnace to 600°C
Stainless steel sieves of 3-in. diameter and
0.062-mm mesh
Sonic water bath to take 3-in. sieves

Fine and coarse brushes for cleaning Desiccator Magnetic stirrer Aluminum weighing dishes Thick wall beakers of 400-mL capacity Porcelain, glazed evaporating dishes

After thawing, the wet sediment sample is added to a pre-weighed aluminum dish and the wet weight of sediment recorded ( $W_W$ ). The sediment is then dried overnight at 105°C to constant weight. After cooling, the sample is stored in a desiccator before determining the dry weight of the sediment ( $W_d$ ). Then:

Percent water content = 
$$\frac{W_{\text{w}} - W_{\text{d}}}{W_{\text{w}}} = 100$$

Note that with some sediment samples, particularly those high in organic content, shell gravel, or stones, it may be necessary to remove the >2-mm fraction before proceeding further with the analysis (see Akagi and Wildish 1975).

Dried sediment from the above (ca. 5-25 g, depending on sediment texture: lesser amounts of coarser and larger amounts of finer sediments) are weighed (Wo) and used for 63  $\mu m$  splitting as follows:

Add the weighed sediment to the thickwalled beaker, with 250 mL tap water, 10 mL of sodium hexametaphosphate solution (6.2 g/L of (NaPO<sub>3</sub>)<sub>6</sub>). Break up the clumps of sediment with a glass rod and stir the sediment slurry with a magnetic stirrer. Leave overnight and quantitatively transfer the sediment plus water with excess tap water to a pre-weighed 3-in. sieve - material passing the sieve, particles <0.063 mm and water, can be discarded. To avoid a buildup of sediments in the plumbing system, we recommend catching the fines on a glass-wool filter at the sink outlet. Spray the sieve with clean tap water and then place the whole sieve plus contents in a sonic water bath containing clean tap water. Repeatedly sonicate and discard the bath water until no further fines pass the sieve. If large amounts of organic matter are present, problems with stickiness and clumping of inorganic grains may occur - a step to remove organic carbon with hydrogen peroxide may be necessary (Akagi and Wildish 1975). Dry the sieve and contents, which now contain sand and >2-mm particles, and reweigh. Subtracting the sieve weight from the latter

gives the dry weight of sedimentary particles >0.063 mm  $(W_1)$ . Then:

Percent silt-clay = 
$$\frac{W_0 - W_1}{W_0}$$
 100

A further 2-5 g of dried sediment is accurately weighed  $(W_0)$  and added to an evaporating dish of known weight. The sample is ashed in a muffle furnace for 2 h at 600°C. After cooling, the weight of the remaining inorganic material  $(W_1)$  is determined. Then:

Percent volatile solids = 
$$\frac{W_0 - W_1}{W_0}$$
 100

One possible source of error with this method is that shell gravel may be oxidized and give spuriously high readings. Acidic pre-treatment of the sample can prevent it.

#### PHYTOPLANKTON ECOLOGY

#### PLANT NUTRIENTS

The following dissolved plant nutrients: ammonia, nitrate plus nitrite and total phosphate can be analyzed by colorimetric methods. The methods all require scrupulously clean glassware and, for safety reasons, working under a fume hood and with proper protective clothing. A suitable colorimetric method for silicate is given in Parsons et al. (1984) but has not been used by us.

#### **Ammonia**

The modified phenolhypochlorite method of Solorzano (1969) was followed. The glassware used with this method must be chemically clean (warm dilute HCl and rinsing with distilled water). The following apparatus and reagents were required:

Spectrophotometer with 10-cm pathlength cuvettes (Beckman scintillation vials - 15 mL capacity)

- A. Phenol alcohol solution. Dissolve 10 g of reagent grade phenol in 100 mL of 95% V/V ethanol USP.
- B. 0.5% sodium nitroprusside. Dissolve 1 g of sodium nitroprusside in 200 mL of

distilled water. Store in amber bottle for up to 1 mo.

C. Oxidizing solution. Mix 100 mL of sodium citrate solution (100 g trisodium citrate and 5 g NaOH in 500 mL distilled water) and 25 mL of hypochlorite solution. Any oxidizing solution remaining should be discarded at the end of the day's analysis.

After cleaning the scintillation vials, they are stored containing equal volumes of solutions A, B and C and a total of ca. 10 mL. At sampling, the mixed reagents are decanted, followed by two rinses of deionized water, then two of sample seawater. To a 15-mL sample of seawater, 0.6 mL of A is added and mixed, a screw cap added and the sample left at room temperature until analysis. Add 0.6 mL of B and 1.5 mL of C, mixing thoroughly after each addition. Leave for 1 h for the color to develop and read the absorbance at 640 nm in a 4-5 cm pathlength cell.

The concentration is determined from a standard curve prepared by freshly weighing 0.100 g ammonium sulphate and making up to 1 L with deionized water. This solution is stable if 1 mL of chloroform is added and stored in a brown glass bottle in the refrigerator. Then:

 $\mu$ g/L - molecular weight of NH $^{-}$ 3 =  $\mu$ M/L 100  $\mu$ g/L  $\div$  19 = 5.26  $\mu$ M/L

Precautions to keep the spectrophotometer cell clean should include limiting use to this analysis and, after cleaning, blanks should be less than 0.01 A for deionized or oceanic seawater. The analysis should be completed as rapidly as possible to avoid loss of ammonia. During analysis, do not use any other ammonia compounds in the lab. If zooplankters are present at high density, the seawater sample should be prefiltered on a suitable size plankton mesh.

#### Nitrate and nitrite

The method used is described in Strickland Parsons (1968)and involves diazotizing determining nitrite after with sulphanilamide and coupling with N-(1naphythyl)-ethyl-euendiamine to form an azo dye whose extinction is measured at 543 nm. In a separate sample, the nitrate is reduced in a column containing cadmium filings coated with copper, to nitrite. Because summer samples contained little nitrite, the first determination was omitted for these samples. Apparatus and stock solutions required are as follows:

Spectrophotometer with 1- and 10-cm pathlength cuvettes
Reducing columns
Plastic sample bottles
50-mL graduated cylinders
125-mL Erlenmeyer flasks

- A. Concentrated ammonium chloride solution. Dissolve 175 g of Anlar NH<sub>4</sub>Cl in 500 mL of deionized water and store in a plastic bottle indefinitely.
- B. Dilute ammonium chloride solution. Dilute 50 mL of A to 2 L with deionized water.
- Cadmium-copper filings. Cadmium filings are obtained from **BDH** Chemicals. Sieve the filings to remove particles smaller than 0.5 mm. Stir about 100 g of filings in 500 mL of 2% W/V copper sulphate (CuSO<sub>4</sub> 5H2O) until the blue color leaves the solution. Put a small plug of glass wool at the bottom of the reduction Fill column with dilute column. ammonium chloride, add the cadmium -copper filings. Filings should be submerged beneath the liquid at all times. ammonium chloride to wash the filings. Flow rate should be between 8-12 min for 100 mL. The filings need to be replaced when the standard values change. Cadmium-copper filings can be reactivated by washing them in a 5% V/V hydrochloric acid solution and then rinsing with distilled water until the pH of the decanted solution is >5. The filings are then added to the copper sulphate solution and the process repeated.
- D. Sulphanilamide solution. Dissolve 5 g of sulphanilamide in 50 mL concentrated HCI (specific gravity of 1.18) and 300 mL deionized water. Make up to 500 mL and store indefinitely.
- E. N-(-1-naphthyl)-ethyleuendiamine dichloride solution.

For the nitrite determination, a 50-mL sample of seawater is added to a flask followed by 1.0 mL of D. After 2-8 min and no further color development, 1.0 mL of E is added. After 10 min but in less than 2 h, measure the extinction at 543 nm in 1-cm pathlength cells. The concentration of nitrite is read from a prepared calibration graph by plotting absorbance at 543 nm versus sodium nitrite concentration. The standard is prepared by drying a small amount of sodium nitrite at 110°C for 1 h. Then, 0.345 g is weighed out and added to 1 L of deionized water and 1 mL chloroform. The stock nitrite solution is stable for months if stored in a dark container. Blanks are run using deionized water to test for reagent purity. Turbidity checks are made by randomly subsampling seawater samples and measuring absorbance at 543 nm against the reagents. The average value is subtracted from the absorbance reading to correct for turbidity.

For determining nitrates, a 100-mL seawater sample is mixed with 2 mL A in a fume hood and added to the reduction tube. The first 40 mL eluted is discarded, but the next 50 mL collected and treated as for nitrite described above. The concentration is determined from a standard curve prepared from a solution containing 1.02 g KNO<sub>3</sub> per L of deionized water. The standard is stable if stored in a dark bottle.

Fresh seawater samples for the determination of nitrate and nitrite may be stored in a refrigerator for 2-3 d but, for longer storage, should be frozen at ca. -20°C. It is necessary to prefilter seawater if the phytoplankton density is high. The sampling bottles must be kept clean by washing in 10% HCl after use and rinsing with deionized water.

#### Total phosphate

Phosphate is determined by the Menzel and Corwin (1965) method. It involves complete oxidation of phosphate and colorimetric measurement at 690 nm. The following apparatus and reagents are required:

Spectrophotometer and 10-cm pathlength cell Autoclave 125-mL Pyrex flasks

- A. 5% potassium persulfate solution. This is prepared fresh daily by dissolving 5 g K<sub>2</sub>S<sub>2</sub>O<sub>8</sub> in 100 mL deionized water.
- B. Mixed reagent. Consists of 100 mL ammonium molybdate solution (15 g ammonium paramolybdate in 500 mL this is stable if stored in a dark plastic bottle) plus 250 mL dilute H<sub>2</sub>SO<sub>4</sub> (140 mL concentrated H<sub>2</sub>SO<sub>4</sub> in 900 mL after cooling, store indefinitely in a glass bottle), 100 mL ascorbic acid solution (27 g ascorbic acid in 500 mL deionized water. Must be kept frozen between uses) and 50 mL of potassium antimonyl tartarate (0.34 g in 250 mL of deionized water a stable solution). The mixed reagent is only stable for 6 h.

To 50 mL of seawater sample, add 8 mL of A in a Pyrex flask and autoclave at 15 psi for 1/2 h. The autoclave is kept in the fume hood during use and is drained and thoroughly cleaned after use to avoid rusting. After cooling the sample, it is made up to 60 mL with deionized water. After standing for 5 min to 2 h, the absorbance is read at 690 nm. Blanks are run with deionized water replacing the seawater sample and the concentration determined from a standard curve. A stock solution is prepared by adding 0.816 g of ovenanhydrous potassium dihydrogen phosphate at 105°C for 3 h to 1 L deionized water and 1 mL of chloroform. solution is stable if stored in a dark container:

 $\mu$ g/L - mol weight of PO<sub>4</sub><sup>-</sup> =  $\mu$ M/L 816  $\mu$ g/L + 95 = 8.59  $\mu$ M/L.

Nutrient data are expressed as dissolved inorganic nitrogen (DIN) or total phosphorous (TP), inclusive of both inorganic and organic fractions, as follows:

DIN =  $(NO_3$ - +  $NO_2$  +  $NH_4$ +) in units of  $\mu$ M/L

TP =  $PO_4^3$  in units of  $\mu M/L$ .

#### MICROBIAL BIOMASS

Determinations of particulate chlorophyll a by fluorometric method and particulate adenosine triphosphate (ATP) by firefly bioluminescence assay are routinely carried out and can be converted to appropriate estimates of microbial biomass as required.

#### Chlorophyll a

Particulate chlorophyll a is determined fluorometrically by the method of Yentsh and Menzel as described in Strickland and Parsons (1968). The equipment and reagents required include:

Turner fluorometer fitted with a high sensitivity door, UV lamp, excitation filters of 5-60 and emission filters of 2-64
1-cm pathlength cuvettes
Millipore filtration equipment inclusive of a small vacuum pump
Pestle, mortar and clean sand
Graduated test tubes and parafilm
Centrifuge

#### A. 90% acetone

B. Magnesium carbonate suspension. Add 1 g of finely powdered magnesium carbonate (reagent grade) to 100 mL deionized water. Shake vigorously just before use.

Add the seawater sample (150 mL to 2 L depending on the chlorophyll content) to a Millipore filter system fitted with a 4.7- or 2.2-cm Whatman GF/C glass fibre filter. As the sample is being filtered, add a few drops of B. Grind the filter in a pestle and mortar containing a little sand with a few mL of A. Quantitatively transfer the 90% acetone extract to a graduated test tube and after repeated washings with acetone, make up to 10 mL. Store the extracts overnight in the dark and centrifuge at setting 5 for 5 min. Read the fluorometer at the appropriate door setting. Door settings (D) are calculated with pure chlorophyll a standards obtained from Sigma Chemical Co., as follows:

$$D = \frac{C}{R}$$

where R is the fluorometer reading and C is the amount of chlorophyll a determined spectrophotometrically at three wave lengths by the following formula:

$$C = 15.6 E_{665} - 2.0 E_{645} - 0.8 E_{630}$$

and E is the extinction at the appropriate wave length in nm.

The chlorophyll a content is expressed as:

mg chlorophyll a per  $m^3 = D \times R$ .

#### Adenosine triphosphate

The currently used method is an adaptation of the firefly bioluminescence assay utilizing synthetic luciferin from Sigma. The following apparatus and reagents are required:

Biolumat B9500 from Laboratorium Prof. Dr. Berthold, D-7547 Wildbad 1, FRG Disposable plastic cuvets Continuously adjustable pipet (0-200 µL) Pipetman P200 Source of good quality, 0.45-µm filtered deionized water Sand bath Hot plate or Coleman stove Graduated boiling tubes with screw caps and 15-mL capacities Millipore Swinnex filter holders fitted with 25-mm diameter, 0.45-µm membrane filters, enclosed in aluminum foil which have previously been autoclaved Disposable, asceptic, plastic syringes 0.02M Tris buffer. Add 2.4228 g Tris salt (Fisher purity of 99.85%) in 1 L deionized water. Adjust pH to 7.8 by adding ca. 2.5 mL 5NHCI Sigma ATP assay mix - add 20 ml, 0.45-µm filtered, deionized water to one vial Fine forceps

Seawater samples are drawn in a syringe and filtered through the Swinnex holder, to which it is attached by means of a luer lock, into a graduated boiling tube until 10-15 mL have been collected. The filtrate is discarded and the dry filter removed with forceps and placed in the boiling tube containing boiling Tris solution heated in the sand bath. After boiling for at least 1 min, the filter is removed and discarded. On cooling, the volume of extracted ATP is measured and, if necessary, stored at -20°C until analysis. The analysis involves luciferin-luciferase in the Sigma ATP assay mix (add 20 mL filtered deionized water to one vial of assay mix) which is injected automatically from the Biolumat (0.1 mL) into a 0.1-mL aliquot of thawed ATP extract. Blanks are run by injecting luciferin-luciferase into Tris buffer

solution and a calibration curve is run each day by diluting standard ATP with Tris buffer. The standard ATP was stored in Tris buffer in sterile plastic vials at -20°C until use. After using the standard in preparing a Tris dilution series (with bacteria-free diluent), the thawed ATP was discarded because of the possibility of bacterial contamination. Live microorganisms rapidly utilize ATP. To calculate the amount of ATP determined from the standard curve in the field sample use:

ATP (ng/L) = 
$$(\frac{\text{sample ATP x extract volume}}{\text{volume of seawater filtered}}) x 1.45$$

The 1.45 accounts for the 45% increase in light flash caused by salinity carryover on the filter (Wildish 1976).

Equivalents for ATP data in terms of cellular carbon or dry weight of bacteria and microalgae are:

Cellular carbon = concentration of ATP x 250 (range 91-333)

Dry weight = concentration of ATP x 750 (range 500-1020)

from various authors (see Wildish 1976).

#### PHYTOPLANKTON SAMPLING

Phytoplankton samples are collected at the surface with a bucket at 10 m depth and 1 m above bottom with 1.8- or 5.0-L bottles. previous study measuring integrated water samples between surface and 1 m and 1-10 m showed similar results to those obtained from samples collected from the surface and 10 m (Martin, unpublished). Ideally, sampling should occur weekly from June through September, biweekly in April, May, October and November and monthly for December through March. A 250-mL sample is immediately preserved with 5.0 mL of a solution of formalin:acetic acid (1:1). Subsamples of 50 mL are later settled in Zeiss counting chambers and analysis of species composition and abundance is made using an inverted microscope. All observed organisms are identified, that is to say, all >2-5 microns in maximum length.

A continuing study of the phytoplankton populations in the salmonid mariculture region is desirable in order to aid and possibly anticipate

the occurrence of blooms and assist in providing an early warning to the growers. Although to date there have been no recorded salmon mortalities as a result of phytoplankton blooms in the Bay of Fundy, the industry is still relatively small and in the developmental stages. Increased production may alter the natural supply of nutrients or conditions in the water column and promote conditions favorable for undesirable and harmful species. Additionally, a particular organism may grow to abnormally high concentrations and cause either oxygen depletion in the water, or release substances stressful to cultured fish. Hyper-nutrification might also provide an environment more suited to the growth of microalgae known to produce toxins.

Phytoplankton studies conducted in 1987 and 1988 (Wildish et al. 1988; Wildish et al., unpublished) in the Letang/Bliss Harbour region indicate a high degree of vertical mixing exists with diatoms being observed throughout the water column. Since the dinoflagellates are motile and photopositive, they tend concentrate in the upper and surface waters. Although the mixing provides favorable conditions such as replenishment of nitrates and phosphates to the surface waters for the growth of phytoplankton, the mixing may influence their growth pattern since it may hinder the cells from accumulating in the more illuminated seawater at the surface. Mixing, combined with the strong tidal action, may carry the cells, nutrients, bacteria, and salmonid farm wastes to more sheltered areas or coves more suited to the growth of high densities of phytoplankton.

Prior to our present work, the only other seasonal studies to determine phytoplankton populations in the Fundy Isles region were in the early 1930s (Gran and Braarud 1935; Davidson 1934). These studies were located in the Passamaquoddy and Cobscook Bay areas. Many of the same organisms have been observed in the Letang/Bliss Harbour area. During the present study it is not only important to establish baseline data for the area, but also to observe whether any fluctuation or growth in populations occur, or whether the introduction of a new, possibly toxic, species has occurred.

The present method of analysis for phytoplankton is time consuming and difficult since there is no one key for the identification of species and there is a distinct shortage of personnel trained or familiar with the structures

of the approximately 150 species observed from the Western Isles region of the Bay of Fundy. A key including photographs taken with a light microscope might eventually be feasible to aid the growers themselves in identifying phytoplankters that might pose a threat to their mariculture operations.

Although studies of *Alexandrium* in the Bay of Fundy have found both seed beds and highest concentrations of cells to be located offshore east and northeast of Grand Manan (White and Lewis 1982; Martin and White 1988), little or no attempt has been made to study the major sink areas or the bloom dynamics and migrations of other algal species for this area. To complement these studies, it is essential to obtain a better understanding of the currents, water movements, and exchange and forces governing the Letang/Bliss Harbour system.

One must realize the difficulty in trying to predict or anticipate phytoplankton blooms since a combination of factors such as salinity, temperature, river runoff, rainfall, hours of sunlight, tidal amplitude, nutrients, and wind influence the timing and intensity of blooms, with no obvious dominant factor being involved. Ecosystem modelling of the type planned for determination of salmonid holding capacity requires information on a seasonal basis for which there is at present little local information. examples include: rate of primary production and dark respiration rates of phytoplankton and bacterial respiration rates in seawater. Α more serious lack understanding in mechanisms, for e.g. how additional plant nutrients affect phytoplankton production, may hinder model construction incorporating cause/effect relationships.

### **BENTHIC ECOLOGY**

The presence of salmonid net pens above a sediment can result in increased levels of sedimentation, particularly of waste food and faecal matter from the fish (Fig. 2). The environmental factors which influence how much of this material reaches the sediments directly under the cages depend on the rate of tidal flows, the rate of supply of wastes from the net pens, the depth of the site, the composition, size and behavior of the particulate matter voided, the temperature and salinity of the seawater, as well as the timing and occurrence

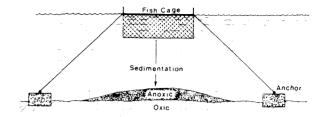


Fig. 2. Diagramatic representation of mariculture sludge buildup under salmonid net pens.

of wind/waves which may result in resuspension and transport of previously settled sediments. Under salmonid net pens where natural sediments are coarse, there may be little buildup of organic wastes because net erosional tidal currents or wave activity control the local Where natural sediments sedimentology. consist of a higher proportion of silt-clay particles and where tidal flows and low wave activity allow net depositional processes to predominate, a buildup of organic-rich material may occur (Fig. 2), which we describe as "mariculture sludge." Frequently this results in development of anoxia within the sediments - a process which has been termed "souring" (Gowen and Bradbury 1987).

A buildup of mariculture sludge results in the loss of aerobic microflora and most macrofauna in the affected area because of the lack of dissolved oxygen or presence of toxic substances in sedimentary pore water. Some species of macrofauna can tolerate anoxic sediments and high sulphide levels by remaining in contact with oxygenated seawater above the sediment, e.g. capitellid polychaetes, and such organisms are resistant to sulphide toxicity.

An anaerobic microbial community replaces the aerobic one in soured sediments. The metabolism of the anaerobic microbes generally falls within one of three major categories: nitrate reduction, sulphate reduction or methanogenesis (Table 1). Because of the heterogenous nature of natural sediments, all three anaerobic respiratory processes may occur simultaneously in microniches, sometimes with concurrent aerobic microbial respiration. The dominant microbial process should be indicated by Eh measurement as shown in Table 1.

One of the purposes in this section is to present the details of Eh sampling because it is

Table 1. Major aerobic and anaerobic respiratory processes found in marine sediments as indicated by Eh measurements (modified from Lynch and Poole, 1979).

Туре	Electron acceptor	Products	Eh, mV
Aerobic	O <sub>2</sub>	H₂O	>0
Anaerobic denitrification	NO <sub>3</sub> ·	NO <sub>2</sub> <sup>-</sup> , NH4 <sup>+</sup> NH <sub>3</sub> , N <sub>2</sub>	0 to -150
sulphate reduction	SO <sub>4</sub> 2-	H₂S, HS	-150 to -200
methanogenisis	CO <sub>2</sub>	CH₄	-250 to -300

a method which has the potential for defining the type and extent of sludge buildup. Nevertheless, calibration and care of the redox electrode is sometimes difficult and interpretation of the data may be problematical. Another purpose here is to present a background and further individual benthic ecological research required to meet the environmental research strategic plan of determining the holding capacity of suitable salmonid culture areas within the Bay of Fundy.

#### Eh MEASUREMENTS OF SEDIMENTS

Like the methods described previously for defining the areal extent of mariculture sludge buildup, such as determining sediment water, silt-clay or volatile solids content, Eh measurement is also a nonspecific indicator of organic addition. Its advantage is that it can help to reveal the major type of microbial community involved by the degree of anaerobiosis which is present.

Ideally the sediment samples should be taken by hand-held corer by a SCUBA diver, but alternatively taken remotely by a Kajak corer. This apparatus is allowed to free-fall into the sediment and generally only takes a satisfactory sample in soft, silt-clay sediments. Sediment samples which do not have a relatively undisturbed sediment-water interface should be discarded before they are used for subsampling. After sampling, the mouth of the corer is partially closed by spreading of the plastic "egg shell" catcher and the upper opening by a messenger-operated rubber plunger. In sampling silt-clay sediments, the

latter was found to be unnecessary and was not always used. The equipment and supplies required include the following:

Dinghy or Eastporter equipped with a motor-driven winch for hauling the corer which is weighted to 20 kg

Core liner tubes drilled in a spiral pattern at 5-cm intervals to take the Eh probe (Fig. 3)

Plastic "egg shell" catchers to retain the sediment sample in the core liner tube

Specific ion meter - Model 407 from Orion with combination redox electrode, Model 96-78

Orion reference electrode filling solution - #900001.

The combination redox electrode must be frequently and routinely calibrated using the method described by Orion. Prepare solutions A and B as follows:

- A. 0.1M K<sub>4</sub>Fe(CN)<sub>6</sub>.3H<sub>2</sub>O, 0.05M K<sub>3</sub>Fe(CN)<sub>6</sub>. Weigh out 4.22 g of the former and 1.65 g of the latter and make up to 100 mL with deionized water.
- B. 0.01M K<sub>4</sub>Fe(CN)<sub>6</sub>3H<sub>2</sub>O, 0.05M K<sub>3</sub>Fe(CN)<sub>6</sub>, and 0.36M KF.2H<sub>2</sub>O. Weigh out 0.42 g of the first, 1.65 g of the second and 3.39 g of the last reagent and make up to 100 mL with deionized water. The expected potential at 25 C depends on the combination redox electrode filling solution as follows:

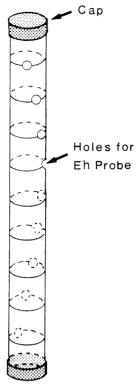


Fig. 3. Core liner tube made from a tube 5 x 50 cm long and holes drilled in a spiral pattern at 5-cm intervals.

	<u>#900011</u>	<u>#900001</u>
Solution A	+234 mV	+194 mV
Solution B	+300 mV	+260 mV

Because of the low salt content in seawater, we have always used the weaker electrolyte filling solution (#900001) and, for convenience, present the expected and observed potentials at different temperatures for this (Table 2, 3). The data of Tables 2 and 3 were obtained using a Lauda K-21 RD recirculating water bath for temperature control. Regression constants for all three probes in solution A (Table 2) are: Eh value = -1.705 (temperature, °C) + 239.571,  $R^2 = 0.96$ , N=21. If the mV readings vary by more than 15 mV from the expected value (Table 2, 3), the difference is added if the A and B values are lower, and subtracted if they are higher than in Tables 2 and 3. electrode should be cleaned by rinsing with deionized water and blotting with absorbant paper, scrubbing the platinum electrode surface with dental floss and an abrasive surface and, if disagreement with calibration mV values is marked, changing the Eh probe filling solution.

The mV readings obtained from field collected samples should be expressed relative to the normal hydrogen electrode (NHE) by:

$$E_{NHE} = E_0 + C$$

where E<sub>0</sub> = mV of unknown and C = mV of reference relative to NHE (Table 4).

#### BENTHIC MICROFLORA

Although it is probably not necessary to know the taxonomic identity of each species of microorganism involved in aerobic or anaerobic metabolism sediment for most practical purposes, it is sufficient to know the proportion of each physiological type present, or at least its contribution to the final gaseous products produced (Table 1). Commonly recognized forms of respiratory processes among bacterial microflora include three types of nitrate reduction: denitrification, nitrate respiration, and The latter dissimilatory nitrate reduction. process may be the dominant one under salmonid farming conditions (Kaspar 1982: Kaspar et al. 1988), apparently because the high level of nitrate in fish farm wastes inhibits denitrification and nitrate respiration. Possible ways to determine physiological types present involve gas sampling and analysis (see Samuelsen et al. 1988) and measurement of biochemical activities in sediments acetylene inhibits dissimilatory nitrate reduction -Kaspar 1982).

Knowledge of the type of sedimentary microbial community metabolism occurring allows a prediction of the chemical oxidation rates across the sediment-water interface. Biological and chemical oxygen demands can also be measured directly and specifically under net pen sites using enclosed sediment gas can chambers which be sampled predetermined time intervals (Hargrave and Phillips 1981). Chemical oxygen demands are determined in a separate chamber in which the microorganisms have first been killed by the addition of a toxic agent. Other methods to do this include miniature redox probes which are used to profile sediments at intervals of 1 mm or so.

Table 2. Expected and observed potential in mV for three different probes in solution A at various temperatures with Orion electrode filling solution 900001.

			Observed			
Temp, °C	Expected	Probe 1	Probe 2	Probe 3		
5	230	231	225	225		
10	221	228	221	222		
15	212	223	214	215		
20	203	209	206	208		
25	194	198	195	195		
30	185	192	188	187		
35	176	181	175	177		

Table 3. Expected and observed potential in mV for three different probes in solution B at various temperatures with Orion electrode filling solution 900001.

			Observed	
Temp, °C	Expected	Probe 1	Probe 2	Probe 3
5	296	285	281	280
10	287	282	275	275
15	278	273	267	268
20	269	262	255	258
25	260	257	248	250
30	251	245	230	242
35	242	235	228	232

Table 4. Reference electrode potential in mV relative to NHE (C) for different temperatures and filling solution concentrations, from Orion literature.

Temperature	Orion #900001	Orion #900011
(°C)	1.5M KCI	saturated KCL
5	254	219
10	251	214
15	249	209
20	244	204
25	241	199
30	238	194
35	235	189

# NUTRIENT FLUXES AT THE SEDIMENT-WATER INTERFACE

The rates of nutrient fluxes, particularly nitrogen and phosphorous compounds, depend on the type of microbial community present as well as on many physical/chemical factors. For example, ammonium absorption has been reported to occur by Rosenfeld (1979) in anoxic marine sediments. The question of nutrient cycling between the water column and sediment has been studied by Hartwig (1976), Aller (1980) and Sayles (1979, 1981). Smith (1971) has described a sampler for collecting seawater close to the sediment-water interface.

In order to properly balance the carbon, nitrogen, and phosphorous fluxes which occur in seawater around a net pen site, it is important to be able to measure the biological and chemical fluxes which occur across the sedimentary surface. Our knowledge of this complex subject is far from complete as is a suitable sampling scheme for determining fluxes on a seasonal basis.

#### PELAGIC-BENTHIC COUPLING

The relationship between pelagic and benthic macrofaunal production has recently become a fashionable research issue among biological oceanographers (e.g. Emerson et al. An important link in pelagic-benthic coupling is provided by particulate matter which is produced by pelagic biota and is transported in a variety of ways to the sea floor. Particulate sedimentation can be caught in floating sediment cylinders of a given diameter and aspect ratio, and some seasonal studies have been made near the mouth of the Bay of Fundy (Emerson et al. 1986). The quality and quantity of particulate matter sedimentation is highly variable seasonally and not all of the energy reaching the sea floor by sedimentation is utilized by in situ benthos: some is horizontally transported to other parts of the Bay and some is buried in situ. Because of the energetic tidal and wind/wave activity in the Bay of Fundy, conventional sediment trapping is not feasible in many areas.

We know of no detailed seasonal studies of pelagic-benthic coupling near a salmonid net pen, although some measurements of sedimentation rates have been reported from Norway and Scotland (Hakanson et al. 1988; Gowen and Bradbury 1987).

#### PHYSICAL OCEANOGRAPHY

Development of knowledge of the holding and assimilative capacities of the Letang Inlet system requires integration of the physics, chemistry, biology and sedimentology. Some of the physical oceanographic questions that need to be addressed include: knowledge of the current regime and the driving forces; knowledge of the flushing within the system and the exchange rates with offshore waters; and the dynamics and kinematics of particulate matter. Providing answers to these questions needs to be approached both from a theoretical and experimental viewpoint.

Although there is a relatively long history of marine research in general for the Quoddy Region of New Brunswick (for an overview, see 1983), oceanographic Thomas studies undertaken in the Letang Inlet system are limited (Wildish et al. 1986, 1988; Loucks 1988; Wildish 1988). Many of the earlier studies in Quoddy Region were associated with assessing the environmental impact of tidally generated electric power by controlling flow in Passamaquoddy and Cobscook Bays with a series of dams and gates (e.g. Chevrier and Trites 1960; Forrester 1960; Trites 1961; Trites and MacGregor 1962). Later environmental studies were focused on assessing environmental impact associated proposed refinery at Eastport, Maine (Anon. 1974; Scarratt 1979).

### TIDES AND TIDAL CURRENTS

The tidal range varies from a minimum of about 4 m to a maximum of about 8 m, depending on astronomical conditions. lunar, semi-diurnal tide (M<sub>2</sub>), with a period of 12.4 h, is the principal tidal constituent. there is significant fortnightly spring/neap variation in the tide associated with the beating of the 12.4-h lunar tide M2 with the 12.0-h solar tide  $S_2$ , the more important modulation over a month is associated with the moon's distance from the earth. This effect is shown as an example in Fig. 4 where the change in tidal range between 14 January and 28 January, which is associated with the minimum and maximum distance of the moon from the earth, is greater than the change associated with the phase of the moon.

Although the vertical tides have been measured at a number of sites in the area and

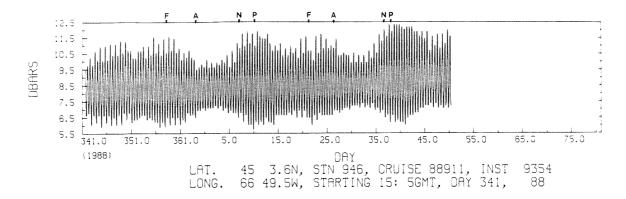


Fig. 4. Water level variation in Lime Kiln Bay during 6 December 1988 (Julian day 341) to 20 February 1989 (Julian day 051) as measured by pressure gauge on moored current meter. N = new moon; F = full moon; A = lunar apogee; P = lunar perigee.

reliable predictions can be made, the same is not true for the tidal currents. The very irregular bathymetry and convoluted coastline result in an extremely complex and variable current pattern. Although strategically placed current meters, moored for not less than a month, should provide valuable information on some of the dominant features of the tidal current, it would be prohibitively expensive to acquire sufficient data to resolve the tidal current patterns on the scale needed in relation to specific aquaculture sites. An alternative is to undertake numerical modelling of the tidal currents, and deploy current meters at a few strategically located sites to validate or calibrate the model.

A numerical model of the Letang Inlet system is being undertaken by ASA Consulting To a first approximation, the large semi-diurnal tides are assumed to be the primary force driving the currents in the area. The model will have three output components. The first will enable one to look at the variation with time in the strength and direction of the tidal currents for any grid-box in the area (ca. 100-200 m scale). The second output will display the tidally driven residual current pattern, and the third, referred to as a water quality model, will enable one to predict the subsequent distribution and concentration of a dissolved or neutrally buoyant "pollutant" introduced at any given point(s) in the area.

One must, however, exercise a healthy scepticism in believing everything the model predicts since the model will only be reliable if

the various simplifying assumptions are valid. For the model currently being developed by ASA, it is assumed that: a) the only driving force is the astronomical tide; b) freshwater input is negligible and the water is vertically and laterally homogeneous; and c) wind stress is negligible. In addition, only very simple representations can be made for bottom friction and diffusive processes.

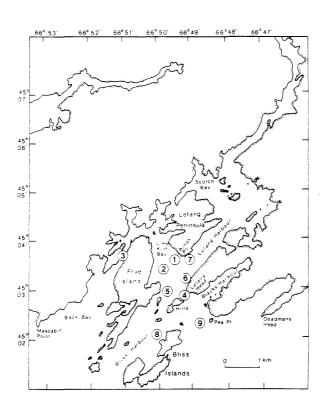
Confidence in the model output requires be verified with selected field measurements of currents and diffusion. In this respect, a current meter mooring program was initiated by DFO in May 1989. During 1989, measurements were made at nine sites in the Letang Inlet system (Fig. 5). The time and duration of each of the moorings are given in Table 5. The validity of the water quality component of the model is dependent on an accurate representation of turbulent exchange coefficients. In this respect, there is only sketchy field data available (Loucks 1988), and further measurements should be undertaken with droque-cluster experiments.

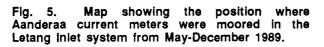
### AREAS, DEPTHS AND VOLUMES

In order to develop preliminary estimates of parameters important in determining holding capacity of the Inlet system (e.g. oxygen supply and demand), a number of computation exercises have been undertaken involving dividing the inlet into segments to determine the low tide and intertidal areas and volumes (Fig. 6). Until further field data and model results become available, we have arbitrarily assumed

Table 5. Periods for which current meters were moored at selected sites (Fig. 5) in the Letang Inlet system, May-December 1989.

Site #	May	June	July	Aug.	Sept.	Oct.	Nov.	Dec.
1					William Control of the Control of th			
2	<del> </del>							
3	-		——					
4							H	
5	-							
6								
7			-					
8								
9					<b> </b>	***************************************		





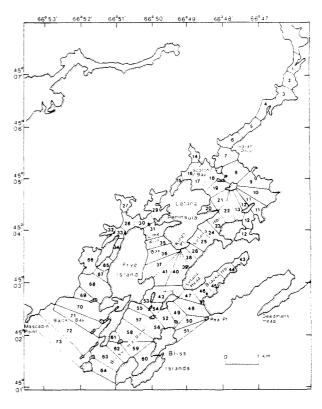


Fig. 6. Map of the 73 subareas of the Letang Inlet system used in computing areas and volumes.

that the Letang Inlet system can be considered as three separate units (Fig. 7): 1) the Letang Estuary (including Lime Kiln Bay); 2) Back Bay; and 3) Bliss Harbour.

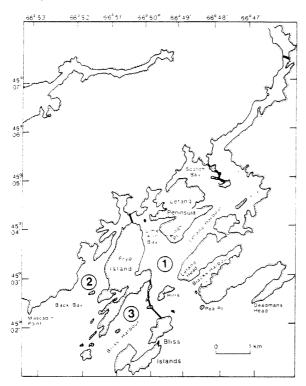


Fig. 7. Map showing the major subdivisions of the Letang: 1) estuary; 2) Back Bay; 3) Bliss Harbour.

Cumulative volumes for each of these areas have been compiled, from which mean tidal excursion can be determined (e.g. Fig. 8). For Lime Kiln Bay, the mean tidal excursions under minimum and maximum tides are These computations displayed in Fig. 9. assume that there is no lateral or vertical variation in current speeds and that the currents flow into and out of the Bay following a simple sinusoidal curve. Although this may not be an unreasonable assumption where Lime Kiln Bay as a whole is considered, it is clearly unwarranted on the scale of a particular A current meter moored near the salmon demonstration and development farm (Fig. 5, site 1) on the northeast side of Lime Kiln Bay from December 1988 until March 1989, vividly displays the difficulty in making such simplifying assumptions. Although the vertical tide displays a relatively smooth rhythmic

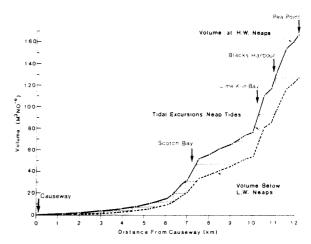


Fig. 8. Plot of cumulative volumes at low and high water neaps in Letang estuary (subdivision 1 in Fig. 7). From this plot, mean tidal excursions during neap tides can be determined for any part of the estuary.

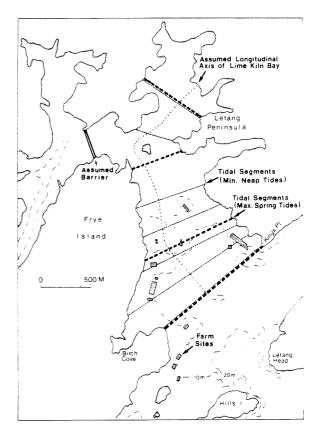


Fig. 9. Tidal segments for Lime Kiln Bay showing mean tidal excursions under maximum spring tides and minimum neap tides.

variation (Fig. 4), the horizontal flow is much more complex, displaying a rotary (rather than rectilinear) feature and with a dominant inward flow (Fig. 10). When averaged over the length of record (2 months), the mean inward flow exceeds 5 cm s<sup>-1</sup>. If this current speed persisted on the northeast side of the Bay, it would displace water more than 1 km over a 6-h period, and exceed the mean computed tidal segment under spring tide conditions by a factor of two! Similarly the M<sub>2</sub> tidal constituent with an amplitude of about 8 cm s<sup>-1</sup> at the mooring suggests a tidal displacement much larger than the mean.

The results from this one mooring strikingly display the complex nature of the currents and point to the need for a numerical modelling approach to develop a picture of the flow patterns in the L'Etang Inlet system.

# DISSOLVED OXYGEN BALANCES IN LIME KILN BAY

The physical characteristics shown in Fig. 8 are used to determine volumes and sediment surface areas for segments 29-37 as shown in Fig. 6. Characteristics of Lime Kiln Bay to determine a budget for the dissolved oxygen are as follows:

- temperature ca. 15°C
- salinity ca. 30 o/oo S
- dissolved oxygen is 100% saturated at the beginning of the night (dark) period

- nighttime lasts for 10 h
- LW occurs at the end of the dark period
- reaeration rates of oxygen across the air/seawater interface are considered to be negligible
- average LW and HW volumes and surface areas calculated
- up to 1/10 of Lime Kiln Bay has become anoxic due to sediment "souring"

### Dissolved Oxygen (DO) Balance

Oxygen available
100% saturation of DO = 5.87 mL/L or
8.39 mg O<sub>2</sub>/L
DO in 1 m<sup>3</sup> = 1000 x 8.39 mg = 8.3857 g

Av. volume = 
$$\frac{LW + HW}{2} = \frac{11.25 + 25.0}{2}$$

 $= 18.125 \text{ m}^3 \times 10^6$ 

 $18.125 \text{ m}^3 \times 10^6 \times 8.3857 \text{ g} = 151,991 \text{ kg}$  DO is available at the beginning of nighttime

#### Oxygen Utilization During Night Time

- microbial respiration in seawater ca. 0.003 g  $O_2/m^3 h^{-1}$  (Kristmanson et al. 1976) .003 x 18.125 x 10<sup>6</sup> x 10 h = 543.80

### Sedimentary Oxygen Demand

The area covered by seawater is estimated from Fig. 6.

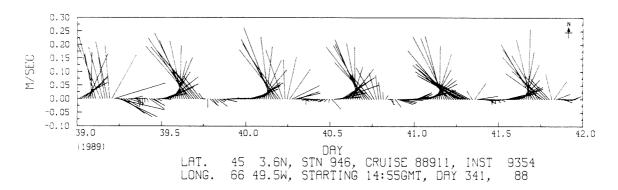


Fig. 10. Stick diagrams of currents at 20-min intervals at site 1 in Lime Kiln Bay for a 3-d period (8-11 Feb. 1989) during spring tides.

Av. area = 
$$\frac{LW + HW \text{ area covered}}{2}$$
 =

$$\frac{1.2 + 2.46}{2} \text{ m}^2 \times 10^6 = 1.83 \text{ m}^2 \times 10^6$$

Oxic sediments - includes 9/10 of available sediment, thus:

1.83 m<sup>2</sup> x  $10^6$  x 0.08 g/m<sup>2</sup>/h<sup>-1</sup> x 10 h x 9/10 = 1318 kg (from Edberg and Hofsten 1975)

Anoxic sediments - for 1/10 of available sediment:

0.183 m<sup>2</sup> x  $10^6$  x 0.5 g/m<sup>2</sup>/h<sup>-1</sup> x 10 h = 915 kg (from Poole et al. 1976)

#### Herring Respiration

557,000 kg of fish respiring 0.45 g O<sub>2</sub>/kg fish/h<sup>-1</sup> (from Henderson, Salmon Growers Assoc., St. George, N.B., pers. comm.; Hakanson et al. 1988)

for 1 h = 
$$250.7$$
 kg  
for 10 h =  $2507$  kg

#### Salmonid Respiration

30,000 market fish = 72,380 kg x 0.4012 g  $O_2$ /kg fish/h<sup>-1</sup> x 10 h = 292.6 kg (from Hakanson et al. 1988 for rainbow trout)

165,000 market fish = 398,090 kg = 1609.1 320,000 market fish = 771,200 kg = 3117.2

#### Balance

151,991 - (545 + 1318 + 915 + 2507 + 3117) = 143,589 or 94.4% remains, and 5.6% of the DO is utilized during the night (Fig. 11).

#### DISCUSSION

A major objective of aquaculture ecology research is to develop a salmonid holding capacity model for a local area which can predict sustainable levels of production. Our feeling is that this model should involve oxygen and/or plant nutrient balances. We point out though that the cause-effect relationship between plant nutrients and increased phytoplankton production is poorly known in the

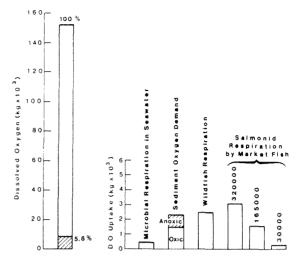


Fig. 11. Dissolved oxygen budget for the night time period (10 h) in Lime Kiln Bay.

marine environment and that an empirical relationship between nutrients and primary production is not available from literature sources, although it is in the freshwater literature. There are also many gaps in locally available biological data, particularly in the seasonal patterns of variables such as primary seawater bacterial production, respiration. pelagic-benthic coupling, sediment-seawater exchanges, and seasonal intensity of wind-wave As reliable data to fill these gaps become available, we would expect a more realistic holding capacity model to be made.

Our research strategy does not only depend on the development of a numerical model with predictive power but also on real-time monitoring of key variables in the mariculture ecosystem as indicators of environmental wellbeing. The variables that should be monitored include the following:

- percentage dissolved oxygen relative to saturation levels
- salmonid mortalities, growth and food conversion rates
- phytoplankton monitoring to determine bloom episodes
- benthic monitoring to determine the incidence and extent of mariculture sludge buildup.

A key element in making these determinations is that they should be available to managers of the industry as rapidly as possible. This is

because they may be required in making decisions - e.g. in siting and expansion or in taking possible remedial measures in the event of a harmful microalgal bloom.

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Appendix 1. Oxygen solubility in mL/L of air in seawater at an atmospheric pressure of 760 mm Hg calculated according to Weiss (Deep-Sea Res., 17: 721, 1970).

	S (o/oo)																	
T (°C)	0	2	4	6	8	10	12	14	16	18	20	22	24	26	28	30	32	34
0	10.22		9.94	9.81	9.67	9.54	9.41	9.29	9.16	9.04	8.91	8.79	8.67	8.56	8.44	8.32	8.21	8.10
1	9.94	9.80	9.67	9.54	9.41	9.28	9.16	9.04	8.91	8.79	8.68	8.56	8.44	8.33	8.22	8.11	8.00	7.89
2	9.67	9.54	9.41	9.28	9.16	9.04	8.92	8.80	8.68	8.56	8.45	8.34	8.22	8.11	8.01	7.90	7.79	7.69
3	9.41	9.28	9.16	9.04	8.92	8.80	8.68	8.57	8.45	8.34	8.23	8.12	8.01	7.91	7.80	7.70	7.60	7.50
4	9.16	9.04	8.92	8.81	8.69	8.57	8.46	8.35	8.24	8.13	8.02	7.92	7.81	7.71	7.61	7.51	7.41	7.31
5	8.93	8.81	8.70	8.58	8.47	8.36	8.25	8.14	8.03	7.93	7.83	7.72	7.62	7.52	7.42	7.33	7.23	7.14
6	8.70	8.59	8.48	8.37	8.26	8.15	8.05	7.94	7.84	7.74	7.64	7.54	7.44	7.34	7.25	7.15	7.06	6.97
7	8.49	8.38	8.27	8.16	8.06	7.95	7.85	7.75	7.65	7.55	7.45	7.36	7.26	<b>7</b> .17	7.08	6.98	6.89	6.81
8	8.28	8.17	8.07	7.97	7.86	7.76	7.66	7.57	7.47	7.37	7.28	7.19	7.09	7.00	6.91	6.82	6.74	6.65
9	8.08	7.98	7.88	7.78	7.68	7.58	7.48	7.39	7.30	7.20	7.11	7.02	6.93	6.84	6.76	6.67	6.59	6.50
10	7.89	7.79	7.69	7.60	7.50	7.41	7.31	7.22	7.13	7.04	6.95	6.86	6.78	6.69	6.61	6.52	6.44	<b>6</b> .36
11	7.71	7.61	7.52	7.42	7.33	7.24	7.15	7.06	6.97	6.88	6.80	6.71	6.63	6.54	6.46	6.38	6.30	6.22
12	7.53	7.44	7.35	7.26	7.17	7.08	6.99	6.90	6.82	6.73	6.65	6.56	6.48	6.40	6.32	6.24	6.17	6.09
13	7.37	7.27	7.18	7.10	7.01	6.92	6.84	6.75	6.67	6.59	6.50	6.42	6.34	6.27	6.19	6.11	6.04	5.96
14	7.20	7.12	7.03	6.94	6.86	6.77	6.69	6.61	6.53	6.45	6.37	6.29	6.21	6.14	6.06	5.99	5.91	5.84
15	7.05	6.96	6.88	6.79	6.71	6.63	6.55	6.47	6.39	6.31	6.24	6.16	6.08	6.01	5.94	5.87	5.79	5.72
16	6.90	6.81	6.73	6.65	6.57	6.49	6.41	6.34	6.26	6.18	6.11	6.03	5.96	5.89	5.82	<b>5</b> .75	5.68	5.61
17	6.75	6.67	6.59	6.51	6.44	6.36	6.28	6.21	6.13	6.06	5.99	5.91	5.84	5.77	5.70	5.64	5.57	5.50
18	6.61	6.54	6.46	6.38	6.31	6.23	6.16	6.08	6.01	5.94	5.87	5.80	5.73	5.66	5.59	5.53	5.46	5.40
19	6.48	6.40	6.33	6.25	6.18	6.11	6.03	5.96	5.89	5.82	5.75	5.69	5.62	5.55	5.49	5.42	5.36	5.29
20	6.35	6.28	6.20	6.13	6.06	5.99	5.92	5.85	5.78	5.71	5.64	5.58	5.51	5.45	5.38	5.32	5.26	5.20
21	6.23	6.15	6.08	6.01	5.94	5.87	5.80	5.74	5.67	5.60	5.54	5.47	5.41	5.35	5.28	5.22	5.16	5.10
22	6.11	6.04	5.97	5.90	5.83	5.76	5.69	5.63	5.56	5.50	5.44	5.37	5.31	<b>5</b> .25	5.19	5.13	5.07	5.01
23	5.99	5.92	5.85	5.79	5.72	5.65	5.59	5.52	5.46	5.40	5.34	5.28	5.21	5.15	5.10	5.04	4.98	4.92
24	5.88	5.81	5.74	5.68	5.61	5.55	5.49	5.42	5.36	5.30	5.24	5.18	5.12	5.06	5.01	4.95	4.89	4.84
25	5.77	5.70	5.64	5.58	5.51	5.45	5.39	5.33	5.27	5.21	5.15	5.09	5.03	4.98	4.92	4.86	4.81	4.75
26	5.66	5.60	5.54	5.48	5.41	5.35	5.29	5.23	5.17	5.12	5.06	5.00	4.95	4.89	4.83	4.78	4.73	4.67
27	5.56	5.50	5.44	5.38	5.32	5.26	5.20	5.14	5.08	5.03	4.97	4.92	4.86	4.81	4.75	4.70	4.65	4.60
28	5.46	5.40	5.34	5.28	5.23	5.17	5.11	5.05	5.00	4.94	4.89	4.83	4.78	4.73	4.67	4.62	4.57	4.52
29	5.37	5.31	5.25	5.19	5.14	5.08	5.02	4.97	4.91	4.86	4.81	4.75	4.70	4.65	4.60	4.55	4.50	4.45
30	5.28	5.22	5.16	5.10	5.05	4.99	4.94	4.89	4.83	4.78	4.73	4.68	4.62	4.57	4.52	4.47	4.43	4.38