ALGAL BIOPOLISHING OF ZINC

FINAL REPORT

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SUMMARY

Periphytic algae grow in mining effluent ponds and streams, characterized by extremes in pH, elevated metal concentrations, and high suspended solids. Periphyton complexes contain metals in significantly higher concentrations than waste water. Therefore periphyton could be utilized as renewable, polishing agents. The objective of this work was to identify the processes which related periphytic algae to zinc removal from the waste water.

An abandoned polymetallic mine in central Newfoundland has a number of circum-neutral waste waters containing 20-43 mg/L zinc. Six serial polishing ponds (approx. 40 m^3 each) were built in the summer of 1989 receiving a flow ranging between 2.1 L/min to 11.4 L/min. Alder branches in each pond provided about 100 m^2 surface area for growth of algae. The residence time of water in the ponds was between 16 and 80 days.

Significant removal of zinc from the water began several months after the growth surface was provided. At the end of the first growing season a 40% zinc removal rate was recorded. In the middle of the second growing season, 86% removal of zinc was evident, which remained the same in the third season when the highest recorded removal rate was 90%.

Periphyton complexes (algae and precipitate) grew in the first year to approximately 0.2 grams dry weight (gdw) of complex per gram dry weight of alder branch. In the second year, periphyton complex biomass increased to 1.8 gdw gdw^{-1} branch), and in the third year, a further increase to 6.5 gdw gdw^{-1} was achieved. It can be concluded that, as periphytic algal complexes establish, more surface area becomes available and polishing capacity is increased.

However, the periphyton fraction of the complex (L.O.I. at 500° C) can vary widely, ranging from 63% to 13%. Periphyton complexes (both organic and inorganic material together) contained an average of 8% zinc and 11% iron (maximum iron concentration was 26%).

By analyzing the periphyton and precipitate fractions of the complexes, it was determined that theoretically "clean" periphyton could ad/absorb about 10,000 μg/gdw of dissolved zinc. Pure precipitates, on the other hand, could contain between 2.5 and 15% zinc, although extrapolation of the data boundary suggested that precipitates with as much as 50% zinc were possible.

To arrive at design parameters for scale-up of the process, an artificial frame with a quantifiable netting and branch area (together with a bag for collecting sloughed biomass) was used to determine growth in waste water, which between July and August, was determined in the polishing ponds to be 0.73 gdw m^{-2} substrate d^{-1}.

Fertilizer was added to polishing ponds 3 and 5. Growth rates in pond 3 were over twice those in pond 1. Photosynthetic rates of periphyton complexes were measured in the field. The highest photosynthetic rates were produced by periphyton complexes growing in polishing pond 3 (0.95 mg O_2 gdw^{-1} h^{-1}) and the lowest rates were recorded in the open pit (0.03 mg O_2 gdw^{-1} h^{-1}). It can be concluded that zinc removal efficiencies are related to growth rates.

RÉSUMÉ

Les algues périphytiques poussent dans les courants et les bassins d'effluents, caractérisés par des extrêmes dans le pH, des concentrations de métal élevées, et de nombreux solides en suspension. Les complexes de périphyton contiennent des métaux à des niveaux de concentration significativement plus élevés que les eaux usées. Donc, le périphyton pourrait être utilisé comme agents d'épuration renouvelables. L'objectif de ce travail était l'identification des procédés qui liaient les algues périphyliques à l'élimination du zinc contenu dans les eaux usées.

Une mine polymétallique désaffectée dans le centre de Terre-Neuve possède de nombreux bassins d'eaux résiduaires circum-neutres contenant 20-43 mg/l de zinc. Six bassins d'épuration en série (d'environ 40 m^3 chacun) ont été construits au cours de l'été 1989, recevant un débit allant de 2,1 l/min. à 11,4 l/min. Des branches d'Aulne dans chaque bassin procuraient une zone de surface d'environ 100 m^2, pour la croissance des algues. La durée de séjour de l'eau dans les bassins était de 16 à 80 jours.

Une élimination significative du zinc, de l'eau, débuta quelques mois après que la surface de croissance ait été fournie. Au terme de la première saison de croissance, un taux d'élimination du zinc de 40% a été enregistré. À mi-saison, au cours de la deuxième saison de croissance, il était évident que 86% du zinc avait été éliminé, et cela ne changea pas au cours de la troisième saison, lorsque le taux le plus élevé d'élimination fut de 90%.

Les complexes de périphyton (algues et précipité) ont augmenté au cours de la première année jusqu'à environ 0,2 grammes de poids sec (gps) de complexe par gramme de poids sec des branches d'Aulne. Au cours de la deuxième année, la biomasse du complexe de périphyton a augmenté de 1,8 gps gps^{-1} de branche), et au cours de la troisième année, une augmentation supplémentaire jusqu'à 6,5 gps gps^{-1} a été atteinte. On peut ainsi conclure que, lorsque les complexes d'algues s'établissent, une plus grande zone de surface devient disponible, et la capacité à épurer est accrue.

Cependant, la fraction de périphyton du complexe (Perte par calcination à 500°C) peut varier de manière importante, allant de 63% à 13%. Les complexes de périphyton (matériau organique et inorganique ensemble) contiennent une moyenne de 8% de zinc et 11% de fer (la concentration maximale de fer fut de 26%).

Par l'analyse du périphyton et des fractions de précipité des complexes, il a été déterminé qu'un périphyton théoriquement «propre» pouvait ajouter/absorber environ 10 000 μg/gps de zinc dissous. Les précipités purs, d'autre part, pouvaient contenir entre 2,5 et 15% de zinc, bien que l'extrapolation de la limite des données suggère qu'il est possible que les précipités contiennent jusqu'à 50% de zinc.

Afin d'arriver à des paramètres de conception pour l'extrapolation du procédé, une structure artificielle, possédant une zone de treillis et de branche quantifiable (comprenant un sac pour récupérer la biomasse détachée) fut utilisée afin de déterminer la croissance en eau résiduaire qui, entre les mois de Juillet et Août, a été déterminée, dans les bassins d'épuration, comme étant de 0,73 gps m^{-2} de substrat $jour^{-1}$.

Un engrais fut ajouté aux bassins d'épuration 3 et 5. Les taux de croissance dans le bassin 3 furent plus du double de ceux dans le bassin 1. Les taux de photosynthèse des complexes de périphyton furent mesurés dans le champ. Les taux de photosynthèse les plus élevés furent produits par les complexes de périphyton poussant dans le bassin d'épuration 3. (0,95 $mgO_2gps^{-1}h^{-1}$) et le taux le plus bas fut enregistré dans la fosse découverte (0,03 $mgO_2gps^{-1}h^{-1}$). On peut conclure que les rendements d'élimination du zinc sont liés aux taux de croissance.

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1.0 INTRODUCTION

In Canada it has been estimated that at least 9,000 ha are covered by sulphide tailings, located mainly in the provinces of New Brunswick, Quebec, Ontario, Manitoba B.C., and the Territories (RATS 1984). Given present chemical treatment technology, it has been estimated that cleanup of current problems will require several billions of dollars. Current mining concerns are beginning to budget as much as 20% of gross profits over the next 10 to 15 years toward environmental cleanup. Acid mine drainage and associated heavy metal pollution can create problems for thousands of years.

Controlling the problem of ferric hydroxide sludge formation, acid water, and heavy metals is currently handled by adding lime to the waste water before it leaves the mine property. The lime increases the pH to more natural levels and precipitates the sludges and metals. Lime treatment, by itself, however, does not always produce an effluent which meets government regulations.

There is another way to deal with the problem. Ecological engineering utilizes engineering, geochemical and hydrological information about a given mine to site proper run off channels, minimizing the amount of contaminated water. Also, with this information, new ecosystems are designed and placed which can neutralize acidic waste water and precipitate metals as metal sulphides. Using available materials, carbon and nutrient supplements are provided to the newly created "wetland". At the same time, periphyton and aquatic macrophyte populations are utilized to set up biological polishing systems which can either work within the wetland, or in polishing ponds just downstream.

The use of periphyton as biological polishing agents is not a new idea. The accumulation of heavy metals by plant life in the vicinity of mining sites and ore bodies has been observed by many researchers. Kalin et al. (1990) reviewed the current literature on periphyton and metal sequestration, concluding that biological polishing could be developed into a treatment process. Studies using algae as a treatment process, and which describe design parameters on pilot- or full-scale systems are rare (Gale and Wixson 1979).

The objective of this work was to address some of the fundamental questions relating to the sequestration of zinc by periphyton populations from AMD seepages at Canadian mine sites. The use of periphyton to sequester metals on a pilot-scale and full-scale requires design parameters. The sequestration of metals from the water by periphyton was therefore addressed using six pilot-scale experimental pools, receiving waste water from a gloryhole, at a mine site in Newfoundland (Buchans, ASARCO). This work is part of a larger study to implement Ecological Engineering for the close-out of an entire mine site.

This report summarizes work undertaken to understand the relationship between periphyton population growth and zinc removal, at the pilot-scale. In Section 2 the pilot-scale experimental pools are described, along with zinc removal rates recorded over the last several years. Section 3 details the composition of periphyton precipitate complexes from

Buchans, including the use of the LOI to fractionate periphyton from precipitates. The growth of periphyton precipitate complexes is described, both on untouched alder branches and on artificial surfaces (periphyton traps). Section 5 discusses the interaction between zinc and the periphyton populations. In Section 6, the basis for modelling the growth of periphyton community growth and zinc interactions is described. This is followed by a summary and conclusions in Section 7.

1.1 The Periphyton Communities and the Wastewater

The efficiency of removal of both particulate and dissolved metals depends on the algal community structure and the adaptability of community members to the waste water environment. Periphyton is a loose term encompassing algae, cyanobacteria, moss, and fungi which live together in communities attached to natural and artificial substrates in the aquatic environment. For the most part, therefore, this report will use the general term periphyton.

The Buchans mine site contains a number of different environments which support periphyton populations. Three periphyton communities have been identified in the OEP effluent system: a *Ulothrix*-dominated community which inhabits seepages and pop-ups in the First Meadow; a *Microspora*-and moss-dominated community which is proliferating in the OEP and outflow streams; and a *Microspora*-, moss-, *Achnanthes*-, and *Eunotia*-dominated community in the polishing ponds. A *Ulothrix* community dominates the algal flora at the

Drainage Tunnel, while a *Temnogametum* population thrives in the outflow of the tailings pond. Each of these communities not only tolerates elevated zinc concentrations, but removes dissolved metals and precipitates from water flowing over them.

The primary site for this study was the Oriental East gloryhole (pit; OEP) and its outflow stream. Other sites were: 1) the Oriental West gloryhole (pit; OWP; pH 3.5, [Zn] 30 mg L^{-1}); Drainage Tunnel (DT; pH 6.5; [Zn] 20 mg L^{-1}); and Tailings Pond 2 (TP; pH 7.1; [Zn] 2.4 mg L^{-1}; MAP 1).

The Oriental East gloryhole is circumneutral, with 25 mg L^{-1} Zn (0.38 mM) and 10 mg L^{-1} Al (0.37 mM). Below the chemocline (oxic/anoxic), the water contains 60 mg L^{-1} Fe (1.1 mM), which produces extensive ferric hydroxide precipitate in the outflow, when ferrous iron is oxidized and hydrolysed in the surface waters.

Periphyton growing in waste water streams at Buchans are thriving in solutions with 20-30 mg L^{-1} zinc. This concentration range is high for a large number of periphyton. For example, Spear (1981) reviews the tolerance levels for a number of algal species. He finds that for the most part all freshwater algae studied can tolerate zinc levels only below 1 mg L^{-1}. The exceptions are those algae such as *Chlorella*, *Gongrosira*, *Scenedesmus*, *Plectonema* which can tolerate up to 10 mg L^{-1}.

Not only are some members of the Ulothricales and Zygnemales tolerant to elevated zinc concentrations, they can concentrate it well above ambient water levels (Harvey 1967; Whitton 1970; Trollope and Evans 1976). Other algae, such as bacillariophytes, cyanophytes and phaeophytes have also been shown to be not only tolerant, but accumulators of zinc (Rai et al. 1981; Gadd 1990; Maeda et al. 1990).

1.2 Processes by which Metals Asssociate with Periphyton

Zinc removal from waste water occurs by several processes. First, zinc precipitates or colloids can be sieved from the water by periphyton populations (Gale and Wixson 1979). Secondly, dissolved zinc can be sequestered by periphyton, either extra- or intra-cellularly (Bates et al. 1983; Harrison et al. 1986; Xue et al. 1988). Intracellular mechanisms include sequestration with specialized proteins called metallotheins (Gekeler et al. 1988) or with polyphosphate bodies (Jensen et al. 1982). Zinc is a required nutrient in small amounts, complexing with enzymes (Williams 1983). Extracellular mechanisms include: 1) adsorption of dissolved metals to ionic binding sites on the cell wall surface (Xue et al. 1988), or to binding sites on extracellular polysaccharides (Strong et al. 1982). 2) Metals can precipitate onto cell walls in the form of plating due to interactions between metabolism and dissolved metals (Stevens et al. 1990). 3) Metal precipitation can be enhanced in the region around the periphyton (especially the diffusion boundary layer) due to release of oxygen, uptake of carbon dioxide, nutrient transport which results in changes in either enhanced oxidation or changes in pH which affect the solubility of metals in solution around the periphyton.

Interactions between chemical and biological systems operating in OEP outflow waters are not simple. Zinc concentrations in the OEP are at or near saturation. Any upward change in pH enhances precipitation of dissolved zinc as zinc hydroxide or zinc carbonate. Zinc can also be co-precipitated with iron when the high ferrous iron below the chemocline in the OEP oxidizes as it exits the gloryhole. Thus, changes in temperature, pH, oxidation state, and carbon dioxide concentration all affect the precipitation rate of zinc. These chemical precipitation processes interact with periphyton communities. Periphyton metabolic processes in and around the periphyton affect bulk water pH, oxidation state, carbon dioxide concentration, and indirectly, temperature. Thus, periphyton metabolic processes will affect chemical precipitation processes. The greater the biomass per unit waste water volume of periphyton, the greater will be the metabolic effect on zinc precipitation.

2.0 THE POLISHING PONDS

In June of 1989, six ponds were excavated in the First Meadow to act as biological polishing ponds (MAPS 1,2). Water was diverted from the main OEP outflow stream through the 6 ponds in series. Between August and September 1989, 110 alder cuttings were placed in each of the pools (130 in pond 6) to act as surface area on which periphyton could grow. The ponds are, on average, about 0.6 m deep, with a diameter of 9.2 m. Pond volumes range from 24 m^{-3} (pond 5) to 54 m^{-3} (pond 2). The average volume is 40 m^{-3}.

Throughout 1991, flows through the polishing ponds varied from 0.035 L s^{-1} in July to 0.174 L s^{-1} in August. The average flow was 0.122 L s^{-1}. Calculated residence times varied from 80 days (July) to 16 days (August). The average residence time for the summer was 23 days. Short-circuiting within the ponds was undoubtedly occurring, as actual residence times, based on dye tracer studies in 1990, were on the order of hours rather than days.

2.1. Zinc Removal in the Ponds

Water passing through the ponds has been sampled and analyzed several times each year since the alders were added. These data indicate that zinc is being removed from the waste stream as it passes through the ponds (FIGURE 1). The removal started several months after the cuttings were added to the ponds. Towards the end of 1989, significant biological polishing started as evidenced by a 40% removal of zinc between pond 1 and 6 (November 1989). In 1990, the best removal was 86% of the zinc (September), and in July 1991, over 90% of the zinc was removed from water flowing through the 6 ponds (FIGURE 1). The gradual increase in zinc removal efficiencies was paralleled by the development of periphyton populations (see Section 4).

Copper concentrations in OEP water are 100 fold lower than zinc (0.01 - 0.1 mg L^{-1}), and appear to be less affected by biological polishing agents (FIGURE 2). For instance, the November 1991 sampling showed 78% removal of incoming copper. However, redissolution or release of copper from sediments or periphyton did occur, as for example, in May 1991 (FIGURE 2).

3.0 PRECIPITATE FORMATION/PERIPHYTON INTERACTIONS

Periphyton growing on alder branches were found in complexes with inorganic material. The inorganic fraction of the complex, in this case, was composed of mostly metal precipitates. In order to understand the relationship between metals and periphyton, the periphyton and metal precipitate had to be separated. The obvious choice was to "ash" the samples, providing an estimate of the organic carbon content. The inverse of the ash weight is the loss on ignition (LOI), which is defined as the loss of weight after 30 minutes in a muffle furnace at 500 °C. In the case of periphyton and precipitate complexes, however, the weight loss can be due to combustion of organic carbon, sulphides, carbonates, and possibly degradation of hydroxides in the sample.

Most higher plants contain minerals and inorganic materials that give them an LOI of near 95%. Most algae, on the other hand, typically contain more minerals per unit weight (Larcher 1975). Some periphyton can have low LOIs due to natural calcium deposits which form on the cell walls, for example in the Characeae (see Hutchinson 1975), or diatoms (with silicious frustules). Others, can have LOIs as high as 90%, depending on species and environmental conditions.

A rough estimate of the organic carbon content can be made subtracting the LOI of a sample from 100%. However, as discussed above, clean algae do not have an LOI of 100%. At each mine site, there is a population of "clean" periphyton, which can provide a maximum LOI (usually around 85%). At the same time, the LOI of pure precipitates can be determined. Due to the carbonates, sulphides, and hydroxides present, they, too, will not have a 0% LOI; it was usually between 20-30%. The organic component of an unknown PPC is then bounded by an LOI of 85% on one side, if it is clean periphyton, and 20-30% on the other side, if it is pure precipitate. This relationship is shown in FIGURE 3. By using the maximum and minimum LOI from a given mine site, and linearly interpolating between the two, it is possible to more closely approximate the organic content of the PPC. In PLATES 1 and 2 the differences between "clean" and precipitate-collecting periphyton can be seen.

The cleanest periphyton in Buchans (manually washed Microspora from the OEP) had LOIs around 77%, and precipitates from the OWP had LOIs around 19%. Using the relationship shown in FIGURE 3, an estimate of the percentage of precipitate in PPCs was calculated, by linearly interpolating between these two extremes.

3.1 PPC Composition

PPCs in Buchans waste water are associated with high concentrations of metal precipitates. PPC elemental composition was analyzed by Inductively Coupled Plasma Spectroscopy

(TABLE 1). East Pit PPCs accumulated 26% iron, while Drainage Tunnel PPCs contained only 3% iron. Sulphur was not accumulated to a great degree, varying between 0.4 and 1.4% of dry weight. Zinc, however, ranged from less than 0.1% of dry weight in waste rock pile seepage PPCs, to 8.2% of dry weight in polishing pond PPCs. Zinc distributions appeared to be related to the pH of the waste water. Those waste water bodies with pHs near neutrality, and with saturating levels of zinc, provide the conditions necessary to precipitate zinc as zinc carbonate and zinc hydroxide. Those waste waters with acidic pHs, do not contain appreciable quantities of zinc precipitates, nor do the PPCs contain high zinc concentrations.

PPCs collected from the OEP outflow area, polishing pond 1, and polishing pond 6 were analyzed more thoroughly (FIGURE 4). These locations represent sequential locations along the flow path from the OEP. Differences in elemental composition between the different locations would indicate that different processes are operating.

The only metal that showed more than a ten-fold concentration difference at sequential positions downstream, was Mn. Mg and Al were both 2-3 x higher in the polishing pond PPCs than in OEP PPCs. Si, Fe and P all decreased from 2-6x in PPCs from the OEP to polishing pond 6 PPCs. Zinc concentrations in PPCs from each area remained high, and increased slightly from the OEP to polishing pond 6, indicating that periphyton in OEP waste water tolerate high concentrations of zinc.

TABLE 1: Buchans PPC Composition

LOCATION	TAXA	Algae %	Fe %	S %	Zn %	Mn %	Al %	Ca %	Cu %	Other %
Drainage Tunnel	Ulothrix	34.0	3.1	0.6	0.7	-	2.1	0.5	0.2	58.8
Waste Rock Pile	Ulothrix	63.2	19.4	1.3	-	-	0.3	0.3	-	15.5
Tailings Pond	Temnogametum	12.8	4.6	1.2	1.9	0.5	1.0	0.8	0.1	77.1
Meadow	Ulothrix	70.9	4.4	1.4	2.2	0.2	0.6	2.1	-	18.2
Oriental West Pit	Ulothrix	20.5	5.6	0.8	0.3	-	1.2	1.7	-	69.9
Polishing Ponds	Microspora	18.7	10.9	0.5	8.2	1.4	1.2	1.6	-	57.5
Oriental East Pit	Microspora	35.3	25.8	0.4	3.6	0.2	0.5	1.3	-	32.9
Oriental East Pit	Precipitate	29.0*	29.3	0.2	3.7	0.3	0.9	0.6	0.1	35.9
AVERAGES		31.9	12.9	0.8	2.6	0.3	1.0	1.1	0.1	45.7

- denotes percentage smaller than 0.1

* denotes LOI

4.0 PERIPHYTON QUANTIFICATION

4.1 Methods

4.1.1. Branch periphyton

Natural PPC populations were monitored by clipping the end of an alder branch, cleaning off PPCs, and drying both branch and PPC. Mass accumulations were recorded as dry weight of PPC per dry weight of branch.

4.1.2. Peritraps

Peritraps were designed to quantify periphyton growth rates by measuring both growth and sloughing rates. Experimental design also included: 1) estimating the time needed for full development of PPC bio/mass; 2) estimating the effects of fertilizer on periphyton growth. Fertilizer was added directly to the trap, or placed on floats in the surrounding pools. To estimate growth and sloughing processes separately, peritraps were installed in the OWP, OEP, and in polishing ponds 1,3,6.

The traps consisted of an artificial netting structure which housed alder branches which had been freshly cut or submerged in pools for two years. Below the netting was a plastic bag which collected any periphyton falling from the netting or substrates. PPC growth rates could therefore be determined on both netting and alder branches. The same sized netting frame was used at several mine sites for comparative purposes. Substrates (branches), however, were site specific. PPC bio/mass was cleaned off the nets and branches, dried and weighed. Total growth was determined by adding PPC weights on nets and branches to that which had fallen into the bag. The cleaned traps were replaced for regrowth three times during the growing season.

Peritraps were placed in late May 1991. Five traps were placed in each polishing pond, with 6 in each of the gloryholes. Trap 1 contained freshly cut alder branches (new). Trap 5 contained "seasoned" alder branches, i.e. branches that had been submerged in the polishing ponds for 2 years. Some of the traps were cleaned on each successive field trip (#s 1,5), while others were left for two (#s 2,3) or three months before cleaning (#4). Collection and cleaning of traps three times over the summer, gave three growth periods, June, July-August, and September-October.

4.2 Growth of PPCs and Periphyton in Ponds and Gloryholes

4.2.1. PPC bio/mass accumulation rates

PPCs were collected from submerged alder branches between May and October each year. FIGURE 5 shows the development of biological polishing capacity (bio/mass) in the polishing ponds from inception through 1991. PPC development increased significantly with time. In 1989, standing bio/masses were less than 0.2 gdw gdw^{-1} branch. In 1990, with the exception of polishing pond 1, the bio/mass was less than 1 gdw gdw^{-1} branch. In 1991, the average bio/mass was between 2 and 3 gdw gdw^{-1} branch.

Populations of periphyton start growing on natural substrates early in the spring, probably at the time of ice break up. Populations accumulate on the substrates until mid summer, at which time some of the material begins to slough off. Sloughing increases throughout the late summer. The effect of this sloughing, along with decreasing growth rates at the end of the growing season, is a reduction in the bio/mass on branches (FIGURE 5).

In 1991, bio/mass growth on branches peaked in August in ponds 1, 2 and 5, and peaked in July in ponds 3, 4 and 6, indicating that the overall peak was sometime between the July and August sampling dates (FIGURE 5).

Natural periphyton communities on branches begin to slough in mid summer. Estimating total biomass production for the summer, or year, is therefore difficult. However, if a starting date at ice break up (late April) is used, then accumulation rates (change in biomass accumulations with time) can be calculated from the data in FIGURE 5. Accumulation rates increase from April to July. That is, accumulations of periphyton on branches were larger with each successive sampling. After July, however, accumulation rates decreased as sloughing increased. By middle September (day 260), sloughing rates became greater than accumulation rates, causing the accumulation rate to fall below zero (FIGURE 6).

Peritrap PPC growth rates during the summer of 1991, from May 30 to July 5, from July 6 to Aug 23, and from Aug 24 to Oct 15 are given in FIGURE 7. These data include PPCs that were cleaned off netting, branches, and found in bags. During June (shaded bars), the growth rates were the lowest of the summer, and ranged between 0.35 to 1.4 gdw m^{-2} (substrate) d^{-1}. This was probably due to the fact that the traps were new in June, and not to poor growing conditions. High growth rates depend on an existing, growing biomass. With new traps and with cleaning the traps each month, this initial biomass is reduced to near zero, thus restricting growth.

During July - August, the biomass of all traps could be quantified at rates ranging from 0.3 to 2.3 gdw PPC m^{-2} (substrate) d^{-1}. Rates fell during the September-October period, ranging from 0.5 to 1.2 gdw PPC m^{-2} (substrate) d^{-1}.

Throughout the summer, growth rates of PPCs in the Oriental West gloryhole (pit;OWP) were lowest of all sites, usually less than 0.5 gdw m^{-2} (substrate) d^{-1} (OWs; FIGURE 7). Even with added fertilizer, biomass accumulations were only about 0.7 gdw m^{-2} (substrate) d^{-1} (OW6; FIGURE 7).

In the OEP, growth rates of PPCs were the highest of any location (OEs; FIGURE 7). Peak biomass was produced in trap 5 at 2.2 gdw m^{-2} (substrate) d^{-1} (OE5; FIGURE 7). The lowest growth rates occurred in June in trap 1 at 0.3 gdw PPC m^{-2} (substrate) d^{-1} (OE1; FIGURE 7). In the polishing ponds, PPC growth varied between 0.4 gdw m^{-2} (substrate)

d^{-1} in pond 1 (PP11; FIGURE 7) in June, to 2.3 gdw m^{-2} (substrate) d^{-1} in pond 3 in July-August (PP35; FIGURE 7).

By analyzing the trap PPCs separately from the bag-caught PPCs, it was possible to calculate the percentage of material which had been sloughed from the trap. These data are presented in FIGURE 8, where an increasing trend in bag-caught PPCs is shown. PPC weights in bags increased from 7.3% in June, to 19% in July-August, and 32% in September-October.

The traps were also used to quantify PPC colonization rates on new alder branches and those which had been submerged for 2 years. These data are presented in FIGURE 9, where no clear evidence for age affects is shown.

4.2.2. Precipitate-corrected periphyton growth rates

The growth rate of the periphyton fraction of PPCs can be calculated by subtracting out the "partitioned" precipitate contribution. FIGURE 10 shows just the precipitate-converted growth rates of periphyton in the polishing pond traps (see Section 3 for conversion specifics). Growth rates of trap periphyton calculated in this way in June were low in all three ponds, and very similar to each other, as expected from the results in FIGURE 7.

Large differences in growth between trap locations were found in the July-August period. During this period, the greatest production of periphyton was in polishing pond 3. The marked differences in production during this period have been attributed to differences in nutrients (see Section 4.3). Average growth during this period was 0.47 gdw (ppt. cor.) m^{-2} (substrate) d^{-1} in polishing pond 3 (3; FIGURE 10).

In September-October, the growth rates of peritrap PPCs in the polishing ponds were, again, similar to each other, averaging 0.4 gdw (ppt.cor.) m^{-2} (substrate) d^{-1}. The similarity between pond periphyton growth in all three ponds indicates that some outside factor, common to all locations was limiting production, probably both light and temperature. Growth rates of periphyton in the Oriental West gloryhole were consistently the lowest, ranging between 0.1 and 0.18 gdw (ppt.cor.) m^{-2} (substrate) d^{-1} (WP; FIGURE 8). Periphyton on traps in the Oriental East gloryhole grew at rates which exceeded polishing pond rates during each measuring period, except for growth in pond 3 during July-August (EP; FIGURE 10).

When growth rates from Buchans peritrap net structures are compared to growth rates on identical structures at another zinc/lead mine in N. Ontario (FIGURE 11), it became clear that periphyton growth rates were roughly similar between these mine sites. This indicates that dissolved zinc concentrations and pH, which vary considerably between locations, are probably not severely limiting periphyton growth. Periphyton growing in Boomerang Lake (OUTF and MPO) at pH 3.5 and zinc concentrations around 7 mg L^{-1} had very similar growth rates to the OEP, PP1, and DECP during the second growth period (July-August), growth rates to the OEP, PP1, and DECP during the second growth period (July-August), which all had pHs between 6 and 7. The periphyton population from Decant Pond (DECP) was dominated by cyanophytes growing in a tailings pond (pH 7, [Zn] 2 mg L^{-1}). This comparison suggests that a factor other than pH may control periphyton growth.

4.3. Photosynthesis - Biomass/Growth Estimates

As discussed in the previous section, growth of periphyton can be measured by determining dry mass produced on clean surfaces at various times over the growing season. In waste water streams, biomass can also accumulate large amounts of precipitate as shown by low LOIs. Since the current method of calculating the periphyton contribution to PPCs is based solely on LOI, other estimates of periphyton growth were initiated.

The accepted alternate method for productivity estimates involves calculation of carbon fixation rates. This can be accomplished by calculating the loss of carbon dioxide, removed during photosynthesis, from the waste water as it passes over the periphyton, or by measuring the increase in oxygen, produced during photosynthesis.

Photosynthetic rates of PPCs from polishing ponds, Drainage Tunnel, Tailings Pond, the OWP and OEP were quantified in August by measuring the production of oxygen in closed bottles, containing a known amount of PPC. Oxygen concentrations before and after incubation were measured using an oxygen electrode (YSI 54). The method is described by Dor and Levy (1987). Rates were also measured in the lab, under controlled conditions of temperature and artificial light (daylight florescent lamps).

TABLE 2: Light/dark bottle experiments, Buchans

Location	Temp. (°C)	NPP	R mg O_2 gdw^{-1} h^{-1}	
OEP Outf	15	0.03	0.11	cloudy
PP 1	19	0.38	0.67	cloudy
PP 3	16	0.95	0.92	cloudy
PP 3	16	0.18	1.7	sunny
OWP	18	0.62	2.21	sunny
Drain Tunnel	10	0.51	1.91	rainy
LAB	15	0.34	0.29	artificial
LAB	25	0.49	1.0	artificial

Field measurements of net primary productivity (NPP) and respiration (R) were measured on 3, mostly cloudy, rainy days in August. NPP rates ranged between 0.03 to 0.95 mg O_2 gdw^{-1} h^{-1}. Laboratory measurements ranged between 0.34 and 0.49 mg O_2 gdw^{-1} h^{-1} for roughly the same temperature range (TABLE 2). Since the relationship between carbon dioxide uptake and oxygen evolution is approximately 1:1 on a molar basis, this means that polishing pond PPCs have the capacity to fix carbon at a rate of 0.36 mg C gdw^{-1} (PPC) h^{-1}

(0.49 1.375^{-1}). Over the course of a day during the summer, the sunlight may be saturating for 12 h. This means that about 4.3 g C gdw^{-1} (PPC) d^{-1} can be photosynthetically fixed. From TABLE 3, it can be seen that these rates represent a low estimate when compared to other algal and macrophyte plant groups. However, the photosynthetic rate of PPCs in waste water *in situ* is substantial.

By calculating net photosynthetic rates for just the periphyton fraction of the PPC, considerably higher photosynthetic rates can be projected. PPCs from the polishing ponds averaged 19% periphyton (mean, all PPC analyzed). Thus, the projected net photosynthetic rate for the periphyton fraction is 23 g C gdw^{-1} (peri) d^{-1}. These calculated photosynthetic rates are more similar to those of natural aquatic plant communities summarized in TABLE 3.

Because PPC net photosynthetic rates from Buchans polishing ponds are substantial, and when adjusted for the periphyton component alone, are within normal ranges, there appears to be no major inhibition by; a) dissolved zinc levels in the water, or b) precipitate formation and sieving in the water surrounding the periphyton.

TABLE 3: Literature values for NPP from aquatic plants.

PHOTOSYNTHESIS - OXYGEN	NPP (mg C gdw^{-1} (12 h d)$^{-1}$	
Aquatic macrophytes		
Elodea sp.	32	Westlake 1984
Isoetes sp.	0.67	Westlake 1984
Ceratophyllum sp.	42	Westlake 1984
Periphyton		
Cladophora sp.	90	in Wheeler 1982
Cladophora wrightiana	19.3	in Wheeler 1982
Phaeophytes (range)	27-58	in Wheeler 1982
Rhodophytes (range)	7.2-19	in Wheeler 1982

Another method used to determine photosynthetic rates is to measure the loss of carbon dioxide from the waste water. Carbon dioxide is taken up during the process of photosynthesis. The total inorganic carbon concentration of water can be determined using alkalinity and pH measurements. Thus, by measuring the pH and alkalinity of water flowing into and out of the polishing ponds, an estimate of the amount of total inorganic carbon (TIC) removed from solution can be calculated. Because the concentration difference is measured over a large area, it probably represents a lower boundary to the pond system photosynthetic rate. On July 5 and August 23, 1991 pH and alkalinity of water entering and leaving the polishing ponds were measured (TABLE 4). Total carbon dioxide concentrations were estimated. Calculations of the total weight of branches in the ponds, together with PPC mass accumulations per gdw of branch gave estimates of the total PPC mass in the ponds. Based on these parameters the calculated photosynthetic rate on July 5 was 0.02 mg

C gdw^{-1} (PPC) d^{-1} (for a 12 h day). On August 23, the calculated photosynthetic rate was 0.07 mg C gdw^{-1} (PPC) d^{-1} (for a 12 h day). Adjusting the alkalinity-based productivity to biomass, net photosynthetic rates ranged from 0.1 to 0.4 mg C gdw^{-1} (periphyton) d^{-1}, which were lower than measurements made in light/dark bottles.

Both of these photosynthetic methods, however, can be affected by more than just periphyton photosynthesis. Oxygen changes will be influenced by the ferrous/ferric ratio. Oxidation of ferrous iron consumes oxygen, and much of the iron in the OEP waste stream is ferrous iron. Direct measurement of TIC is also affected by two processes; the precipitation of zinc carbonates, and diffusion from the large pond surface area. Since diffusion and precipitation are both acting to remove TIC, the contribution of photosynthesis to the overall dissolved carbon dioxide concentration is underestimated. If ferrous iron is oxidizing and removing oxygen, the light/dark bottle experiments will also underestimate photosynthesis (overestimate respiration).

Thus, minimum carbon fixation rate estimates were between 0.02-0.07 mg C per gdw periphyton per day. Maximum estimates using short-term oxygen production rates were 22 g C gdw^{-1} (peri) d^{-1}. Growth rates of periphyton can also be used in this comparison. Growth from branch PPCs can be converted to fixed carbon. To make this calculation, growth rates of PPCs from FIGURE 5 (ponds 2 and 3) were made over the June growth period (36 days). Growth rates were calculated using the formula (B2-B1)/(B1•T), where B2 is the biomass at T2, B1 is the biomass at T1, and T is time difference between T1 and

T2. The result is that periphyton in the polishing ponds can fix between 0.7 and 10 mg C gdw^{-1} (peri) d^{-1}, assuming that 50% of periphyton is carbon.

TABLE 4: Carbon metabolism based on changes in pond TIC.

PHOTOSYNTHESIS - ALKALINITY CHANGES			
Location	pH	Alk (mg L^{-1})	CO_2 (mg L^{-1})
July 5, 1991			
Pond 1 IN	6.95	183	40.1
Pond 6 OUT	7.33	50	4.2
August 23, 1991			
Pond 1 IN	6.75	210	72.7
Pond 6 OUT	6.75	118	40.9

4.4. Growth Enhancement in the Ponds

Hutchinson (1975) cites a number of studies which set a limit of about 0.1% of dry weight phosphorus as a baseline for phosphate limitation in periphyton and aquatic macrophytes. If a plant contains internal phosphorus levels above this level, they are P-sufficient; below this internal level, and the population is P-deficient. Phosphorus sufficiency may not only elevate growth rates, but it may also protect plants against elevated zinc concentrations, through localized, external precipitation of zinc phosphate and the internal production of polyphosphates (Rana and Kumar 1974; Jensen et al. 1982).

In May 1991, slow-release fertilizer bags were suspended just below the water surface in ponds 3 and 5; pond 3 received 2 bags (1 kg each), pond 5 only one bag. This fertilizer, Osmocote (NPK 19:6:12) was designed to provide an even release rate over 3 to 4 months at 20 °C. Fertilizer increased periphyton production in the ponds in which it was placed. In July-August, the growth rates of periphyton in pond 3 were over twice that in pond 1 (see FIGURE 10). Periphyton growth in pond 6 was also higher than growth in pond 1. Periphyton were greener and had greater amounts of floating biomass in ponds with, as compared to without, fertilizer.

NPK slow-release fertilizer was also added to certain peritraps in the OWP and the OEP. Phosphorus, as a component of the fertilizer, was used as a tracer, allowing differentiation between populations with and without fertilizer. FIGURE 12 presents the phosphorus content of PPCs in the Buchans area. The mean of a number of PPCs are shown (n is above bar). From these data it is evident that PPC populations in the East pit (OEP) and the Drainage Tunnel (DT) contained high concentrations of phosphorus (0.2%) on a dry weight basis. Populations from the waste rock pile seepages (WRP), tailings pond effluent (TP2), and the Second Meadow seeps (2MD) contained around 0.05% P. The Oriental West gloryhole contained an intermediate P concentration (0.1%). The Oriental East and West gloryholes have measurable dissolved P peaks in June-July each year (1-3 mg L^{-1}), which are probably related to spring turnover. These elevated P levels affect the growth of PPCs in June and July, reducing the need for fertilizer.

Comparisons between fertilized and unfertilized peritraps in the two gloryholes and the polishing ponds reinforce the interpretation that the OEP may be phosphorus sufficient, at least for the first part of the summer (FIGURE 13). Oriental East gloryhole PPCs from unfertilized traps contained more phosphorus than PPCs from fertilized traps, although the differences were not statistically significant. The only populations which responded well to added fertilizer were PPC populations in the OWP. There, the differences between unfertilized and fertilized peritrap PPCs was statistically significant.

5.0 ZINC-PERIPHYTON

When PPCs from a variety of sites around Buchans are analyzed for zinc, and graphed against the percentage of precipitate present in the PPC, the relationship shown in FIGURE 14 was found. From these data it is evident that PPCs can have as little as 0.9% or as much as 15% zinc. However, there do seem to be boundaries on the zinc content of PPCs. The lower boundary would suggest that there is a specific lower limit to the zinc content of the precipitate fraction (1.5%) and periphyton fraction (0.9%). The upper limit for precipitates appears to be around 10 to 15% zinc, and the limit for periphyton around 1.5%.

The effect of the chemical and physical environment (location) on PPC composition can be analyzed by ranking the zinc concentrations in PPCs from highest to lowest (FIGURE 15). Ranking the zinc concentrations of Buchans PPCs from highest to lowest, also separates them by the pH of the waste water in which they live. Thus, the highest zinc concentrations are found in water with the highest pH (polishing ponds and Oriental East gloryhole), and the lowest zinc concentrations are found in PPCs from water with the lowest pH (Oriental West gloryhole, waste rock pile seeps). It appears that zinc concentrations in Buchans PPCs are related to the pH at which zinc is precipitated in saturated solutions. Zinc is precipitated either as zinc carbonate or zinc hydroxide. Precipitation of zinc, in saturated solutions such as Buchans waste water, occurs around pH 7. This suggests that the differences in precipitate quantity should be related to the length of time a given water body, surrounding the PPCs, remains at pH 7 or greater. Thus, by increasing the retention time in areas of high photosynthetic activity, the pH would rise and more zinc would be precipitated (as zinc carbonate). In addition, enriched phosphate concentrations in the lower ponds, due to fertilizer applications, would enhance precipitation of zinc phosphate.

PPCs from the OEP outfall were collected several times over the year (PLATE 1). Each sampling period, PPCs were brought back into the laboratory and a 10 gfw subsample was cleaned of debris, and the periphyton washed in distilled water until no precipitates were found in the wash water. The periphyton and precipitate water were dried, powdered, and analyzed by ICP.

Zinc concentrations in the whole samples (uncleaned), cleaned periphyton, and precipitates are shown in FIGURE 16. These data demonstrate that the zinc in these samples is associated with both the periphyton and the precipitate fractions. The cleaned periphyton, with the exception of August samples (labelled 8), contained between 1 and 2% zinc. The precipitate samples (again without August) contained 2-3% zinc. Samples from August, contained the highest concentration of zinc, suggesting that higher pHs and/or longer residence times were evident at the OEP outfall at that time.

A number of laboratory experiments were carried out to further elucidate the interactions of zinc and algal growth, as described in the next section.

5.1. Laboratory Experiments

As outlined in Section 1, there are several methods by which periphyton populations can sequester zinc. We have explored the probability of enhanced precipitation, and biosieving, in the sections dealing with precipitates. However, dissolved zinc is also sequestered by clean periphyton populations. Zinc is either taken up and stored internally in polyphosphate bodies or proteins, or it is adsorbed to cell wall or polysaccharide surfaces.

To look more closely at the phenomenon of dissolved zinc sequestration, laboratory experiments designed to add dissolved zinc to cultured populations of OEP periphyton were performed. Laboratory experiments were designed to define the relationship between dissolved zinc and cultured OEP algae. In these experiments, dissolved zinc concentrations were monitored over time, both with and without algae.

5.1.1 Methods

Periphyton, cultured from PPCs collected in the OEP and maintained in culture medium (Hargreaves and Whitton, 1976) in the lab, were studied for their ability to remove zinc from both OEP water and prepared growth medium. In these experiments, 1 gram (fresh weight) of cultured *Microspora* was placed in 500 mls of either OEP water or culture medium. The concentration of dissolved zinc was measured periodically (Taylor colorimetric test).

5.1.2. Zn Uptake Experiments

In early experiments it was found that in the absence of algae, dissolved zinc precipitated from bicarbonate-buffered water, at concentrations which were naturally found in the OEP (20-30 mg L^{-1}). Agitation or bubbling enhanced the precipitation process. This presented problems for studies of uptake and adsorption of zinc by periphyton. Short-term uptake studies (hours), however, showed that loss of zinc from water was faster in the presence of periphyton than without. The same phenomenon held true over longer term experiments (days). The periphyton were capable of removing zinc from both prepared growth media spiked with zinc as zinc sulphate, and OEP waste water.

Removal of dissolved zinc at concentrations ranging from an initial 30 mg L^{-1} to 1 mg L^{-1}, appeared to be composed of two phases. The first, rapid, phase appeared to be related to zinc adsorption and ionic exchange with algal cell walls or external carbohydrates. The second phase was much slower, and was linear over the rest of the experiment (several weeks; FIGURE 17).

If, over the short-term, zinc is adsorbed onto periphyton surfaces, then there should initially be a linear relationship between bulk water zinc concentration and adsorbed zinc. Specifically, the greater the external concentration of zinc up to 30 mg L^{-1} (highest tested), the greater number of ionic binding sites on the surface will be replaced with zinc, until all potential binding sites are replaced with zinc. The rate of removal would initially be high, slowing down as more of the sites are filled.

If the rapid loss of zinc from the water (< 1 day) is directly linked with an equivalent gain by periphyton, then the disappearance of a known quantity of zinc from the medium will be reflected in appearance of zinc with the algal mass. For example, if 1 gfw of algae can remove 20 mg L^{-1} from a 500 mL incubation flask, then 10 mg of zinc should be found in association with the algae. This accumulation can also be expressed on a dry weight basis as μg gdw^{-1}, and is then comparable to field observations on zinc accumulation. FIGURE 18 relates the zinc disappearance from media with different initial concentrations of zinc (described in FIGURE 17 and other experiments). By replotting the amount of zinc that disappeared from the medium (as a corresponding gain by algae) against the initial concentration of zinc in the medium, a linear relationship is shown. Interpolation of the curve suggests that, at a water zinc concentration of 15 mg L^{-1}, "clean" algae will rapidly gain zinc to concentrations of about 1.5% (15,000 μg gdw^{-1}). This accumulation is consistent with zinc concentrations found in relatively "clean" periphyton (visibly free of precipitates) from several Buchans sites (see FIGURE 14).

After the initial phase, when the cation exchange process has reached a steady state, there remains significant, though slower, zinc removal capability (FIGURE 17).

Coleman et al. 1971 have examined the uptake of zinc by algal cultures. The data from their experiments suggest that *Pediastrum*, *Euglena*, and *Chlorella* all take up zinc in the concentration range from 1-30 mg L^{-1}. Of the species tested, all seemed to saturate uptake at concentrations between 15 and 25 mg L^{-1}. All species concentrated zinc taken up from the medium. In another study by Bates et al. (1983), the physiology of zinc uptake by *Chlamydomonas variabilis* was studied. Bates et al. incubated *Chlamydomonas* in growth media with a spiked zinc concentration of 0.46 mg L^{-1}. They found that the zinc removed from the medium during an initial 2-3 day period, was readily exchangeable with chelators such as ethylenediamine tetraacetic acid (EDTA), indicating that it was bound externally. After 3 days, the EDTA removable zinc was stable. Transported zinc (i.e. internal) increased slowly over the 6 day experiment. They also concluded that the amount of zinc which could be adsorbed was related to the growth rate of the alga.

In a later study, Harrison et al. (1986) found that the amount of zinc adsorbed to algal cell surfaces was dependent on culture age and growth rate for *Chlamydomonas*. Specifically, exponentially growing cells could adsorb the most zinc. Further, a pH decrease, from 7 to

5, resulted in a reduction in zinc adsorbed. A competition between H^+ and Zn^{2+} was hypothesized. Zinc transport (transmembrane) also dropped with increasing cell age, and decreasing pH (from 7 to 5).

In conclusion, zinc uptake by algae is characterized by a short initial phase of rapid adsorption, associated with a short-term loss of metals from solution. This is followed by a relatively longer phase of zinc uptake, which appears to be a function of growth.

5.2. Zinc Removal by Periphyton in the Field

The amount of zinc removed by periphyton in the field can be estimated by analyzing dried samples of PPCs from peritraps and branches. To do this, PPC weights were multiplied by the closest matching (location and date) ICP-determined zinc concentrations. Although not every peritrap sample was analyzed by ICP, at least one sample from each set of traps was analyzed by ICP. If the PPC growth rates, as defined by peritrap data, are multiplied by the percentage of zinc per unit dry weight, a zinc removal rate can be calculated. This is expressed as mg zinc removed per m^2 of substrate per day by total peritrap bio/mass (FIGURE 19). Growth rates on peritraps were lowest in June (see FIGURE 7), highest in July-August, and intermediate in September-October. If zinc precipitation rates were constant throughout the summer, zinc removed would be independent of growth rate.

However, if zinc precipitation rates were not constant, there should be a relationship between zinc removal and growth rate. Such a relationship is apparent.

Zinc removal rates seemed to be highly correlated with periphyton growth rates in the polishing ponds and the OEP, but, only as measured over a given growth period (FIGURE 20). The higher the growth rate, the greater the zinc concentration in the PPC. The slope of the line represents the rate of zinc removal per unit growth. The highest slopes were represented by growth in the polishing ponds. The next two lower slopes exemplify trap growth in the OEP. Finally, at the bottom of the graph are the OWP data. That the two highest slopes should depict growth in the polishing ponds, is consistent with the idea that conditions in the polishing ponds are more amenable to zinc precipitation. The higher the growth rate, the more the chemical conditions of the surrounding bulk solution are changed.

There also appears to be a relationship between growth period and zinc. For instance, June polishing pond peritrap PPCs were the most efficient at collecting zinc for the least growth rate increment (line 1), while July-August polishing pond peritrap PPCs were less efficient (line 2), and the July-August peritrap PPCs in the OEP (line 3) were even less efficient. Finally, the June OEP peritrap PPCs were extremely inefficient (line 4), increasing zinc concentrations by only 7600 μg gdw^{-1} for an increase in growth rate from 0.4 to 0.7 gdw (ppt. corr.) m^{-2} d^{-1}. Since there was little zinc accumulation in PPCs from the Oriental West Pit, regardless of PPC growth rate, a different process must be operating under acidic conditions.

Regardless of the mechanism, a significant fraction of the zinc loading was removed from the waste stream. Calculations from water chemistry data on Aug 25, and PPC growth data over the preceding 2 months are shown in FIGURE 21. These data indicate that under the conditions of: 1) 16 day residence time; 2) 20 mg L^{-1} Zn in the inflow stream, and 3) 6.75 pH, 88% of the dissolved zinc was removed. Up to 40% of that zinc could be accounted for by PPCs in the ponds.

6.0 MODEL DEVELOPMENT

A model simulating the polishing pond system, PPC growth, and zinc is being developed. The model is intended as a framework to study the important parameters controlling growth and zinc removal. It is also intended as a scale-up tool. The primary objective is to predict the growth of periphyton and the removal of zinc throughout the year. The yearly growth of periphyton will be modelled in a Growth Module (SCHEMATIC 2). Coupled with the growth module is the Zinc Sequestration Module, which will utilize both field and lab data on zinc removal mechanisms and rates. These rates, of course, are tied to periphyton growth and geochemical conditions in the effluent stream. Some of the model subroutines are briefly outlined below.

The Physical Environment Module predicts water temperature and sunlight conditions. Based on Buchans monitoring records, the yearly fluctuations in water temperature at the

OEP outflow have been plotted (FIGURE 22). Yearly sunlight (irradiance) fluctuations for the Buchans area have been estimated from sunshine records, theoretical above atmosphere irradiance, and climatic records (FIGURE 23).

The Chemical Environment Module predicts yearly fluctuations in nitrogen, phosphorus, carbon dioxide concentrations. These are based on normal limnological data gathered from field and lab analyses of the Oriental East gloryhole water.

The Geochemistry Module uses the limnological data and interacts with it. Elevated metal concentrations in the mine effluent water alter the concentration and availability of plant nutrients. The Geochemical Module thus acts like a filter, through which nutrients must pass before they are available to the plants. Plant metabolism can also alter the chemical environment in the bulk solution surrounding community, causing geochemical precipitation of metals and nutrients. This particular aspect of the model is the least understood and is still under development. So far, several of the processes controlling nutrient levels and zinc concentrations have been identified.

The Biomass Module predicts the growth and productivity of periphyton based on inputs from the above two modules. Biomass production can, in turn, alter geochemistry and zinc concentrations. The annual growth production of periphyton is ultimately based on measured growth over the summer (see FIGURES 5,7), and photosynthetic rates measured in the laboratory and field.

7.0 SUMMARY AND CONCLUSIONS

The genera *Ulothrix*, *Microspora*, *Achnanthes*, and *Eunotia*, together with moss protonema and some fungi, grow well in waste water containing 10-30 mg L^{-1} zinc. These same genera are found in mining sites across Canada, and have been recorded from other parts of the world in environments with high concentrations of zinc (Kalin et al. 1991, Whitton 1970).

Periphyton populations at the site are found complexed together with precipitates and inorganic sediments. These periphyton precipitate complexes (PPCs) grow and accumulate in large masses on alder branches in experimental pools throughout the summer. PPCs began growing several months after introduction of alder surfaces. In 1989, the greatest biomass recorded was 0.2 gdw gdw^{-1} of branch. In 1990, the highest recorded biomass was 1.8 gdw gdw^{-1} of branch while in 1991 this rose to 6.5 gdw gdw^{-1} of branch (in a fertilized pond). Bio/mass accumulations appear to have levelled off after two years of growth. This plateau may represent a physical weight limitation, which initiates sloughing.

In 1991, PPC bio/mass on branches increased through July. Bio/mass accumulations peaked on alder branches in July-August. Extrapolation of data suggest that a minimum of 36% of the bio/mass measured in July-August was sloughed to the sediments.

Peritraps in the polishing ponds and pits were used to quantify PPC growth. The highest PPC growth rates were in the Oriental East Pit (July-August - 4.5 gdw m^{-2} substrate d^{-1}), and lowest in the OWP (June - 0.2 gdw m^{-2} substrate d^{-1}). Bag data from peritraps indicate that 32% of PPC bio/mass sloughed during the September-October period.

Fertilizer was added to polishing ponds 3 and 5 in the form of slow-release pellets (NPK 19:6:12). Growth rates of PPCs in pond 3 were over twice those in pond 1. Pond 6 PPC growth was also higher than pond 1 PPC growth. Phosphorus was identified as one of the limiting factors in the growth of Buchans periphyton complexes.

The LOI was used to fractionate the PPC into periphyton and precipitates, the LOI of "clean periphyton" and "pure precipitates" were subtracted from the LOI of an unknown sample. A straight line interpolation between the two extremes defined the percentage of precipitate or periphyton. Periphyton fractions thus calculated represented only on average 32% of the weight of the complexes.

Once the periphyton fraction of the PPC could be determined, it was possible to adjust PPC growth rates to represent just the periphyton fraction. Periphyton growth rates calculated in this manner were highest in polishing pond 3 (0.7 gdw m^{-2} substrate d^{-1}), and lowest were in the OWP (0.1 gdw m^{-2} substrate d^{-1}).

A comparison of periphyton growth on peritrap netting was made between Buchans and another mine site in N. Ontario. Growth of periphyton on peritraps in the OEP was similar to growth on other peritraps in a tailings pond (Decant Pond) and Boomerang Lake (0.6 vs. 0.4 vs. 0.4 gdw m^{-2} substrate d-1, respectively) when averaged over the whole summer. Similarities in growth rates between Boomerang Lake PPCs and Decant Pond PPCs and OEP PPCs under different pHs (3.5 vs. 7) suggest that growth of periphyton may not be affected by pH. Comparisons of growth between Decant Pond and OEP (1 vs. 25 mg L^{-1} Zn) also suggest that metal toxicity is unlikely a significant factor.

Photosynthetic rates were measured on a number of PPC populations around Buchans. The highest recorded net photosynthetic rate was 0.95 mg O_2 gdw^{-1} h^{-1} in polishing pond 3. The lowest rate was found in OEP populations (0.03 mg O_2 gdw^{-1} h^{-1}. The highest rate was converted to 22 g C gdw^{-1} d^{-1}, which is within the range of net photosynthetic rates from periphyton in non-polluted environments. These numbers reinforce the suggestion that the periphyton in Buchans waste water are well adapted to growth under these conditions.

PPCs in Buchans waste water are associated with high concentrations of metal precipitates. East Pit PPCs accumulated 26% iron, while Drainage Tunnel PPCs contained only 3% iron. Sulphur was not accumulated to a great degree, varying between 0.4 and 1.4% of dry weight. Zinc, however, ranged from less than 0.1% of dry weight in waste rock pile seepage PPCs, to 8.2% of dry weight in polishing pond PPCs. Zinc distributions appeared to be related to the pH of the waste water. Those waste water bodies with pHs near neutrality, and with saturating levels of zinc, provide the conditions necessary to precipitate zinc as zinc carbonate and zinc hydroxide. Those waste waters with acidic pHs, do not contain appreciable quantities of zinc precipitates, nor do the PPCs contain high zinc concentrations.

After fractionation of PPCs into periphyton and precipitate, the metal concentrations in each fraction were calculated. Thus, clean periphyton were attributed between 0.9 and 1.5% zinc, while precipitates were attributed a minimum of 1.5% zinc, which agreed with known precipitate zinc content of the OEP (1.9%).

Laboratory studies of zinc uptake by cultured populations of *Microspora* suggest that there are two processes by which algae remove dissolved zinc from water. The first process was rapid, occurring over a matter of hours. Adsorption of dissolved zinc accounted for this removal. Extrapolation of lab uptake studies could account for the distribution of zinc found in "clean" field periphyton samples.

Growth rates of PPCs on peritraps and the corresponding PPC zinc concentrations were correlated, suggesting that growth (and photosynthesis) may result in enhanced zinc precipitation. June polishing pond PPCs contained twice the concentration of zinc per unit of growth rate, as July polishing pond PPCs, which, in turn, contained twice the zinc as complexes in the Oriental East gloryhole.

Zinc precipitation in carbonate-buffered waters occurs around pH 7 and above. Since photosynthesis induces pH increases in polishing ponds, the precipitation of zinc carbonate is enhanced, especially in the water directly surrounding the PPCs. Enhanced precipitation is the probable cause of the elevated zinc concentrations in polishing pond PPCs, and the correlation of zinc content with growth. This pH effect on the precipitation of zinc also explains the lower zinc accumulation ability of PPCs from the waste rock seepage and Oriental West gloryhole, which are both acidic.

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FIG 1: ZINC REMOVAL
Ponds 1 and 6

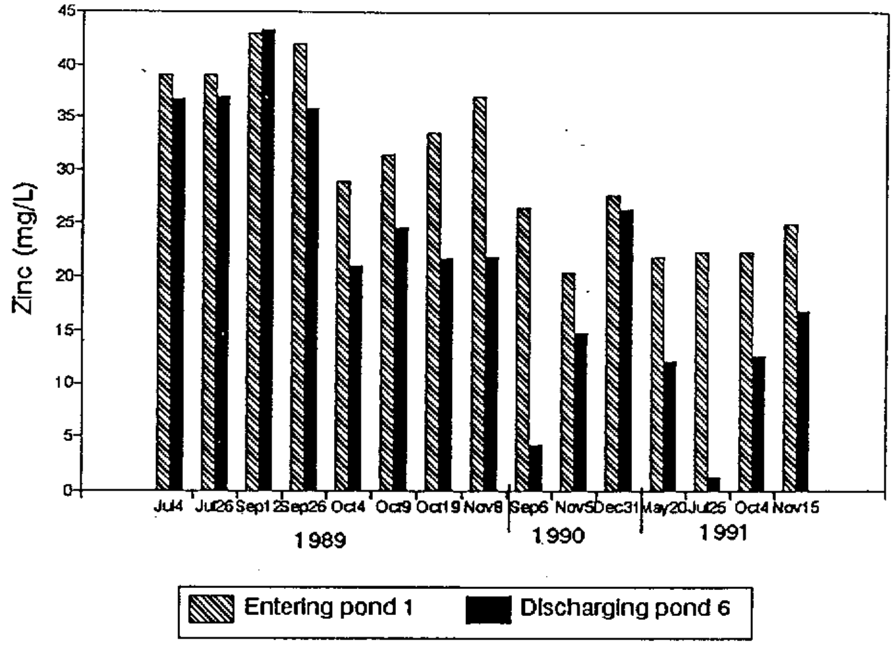


FIG 2: COPPER REMOVAL
Ponds 1 and 6

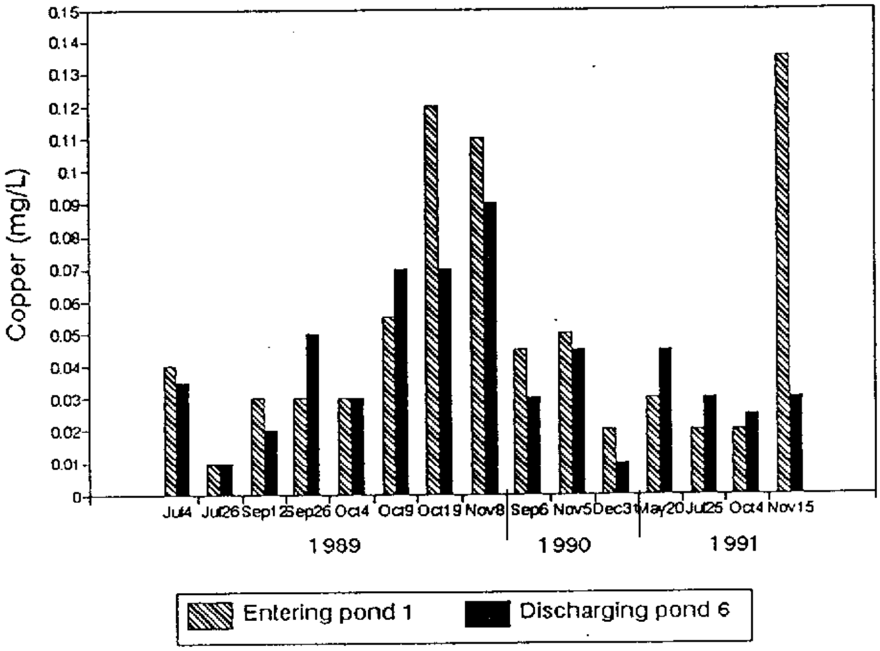


FIG 3: PRECIPITATE vs. LOI
Theoretical

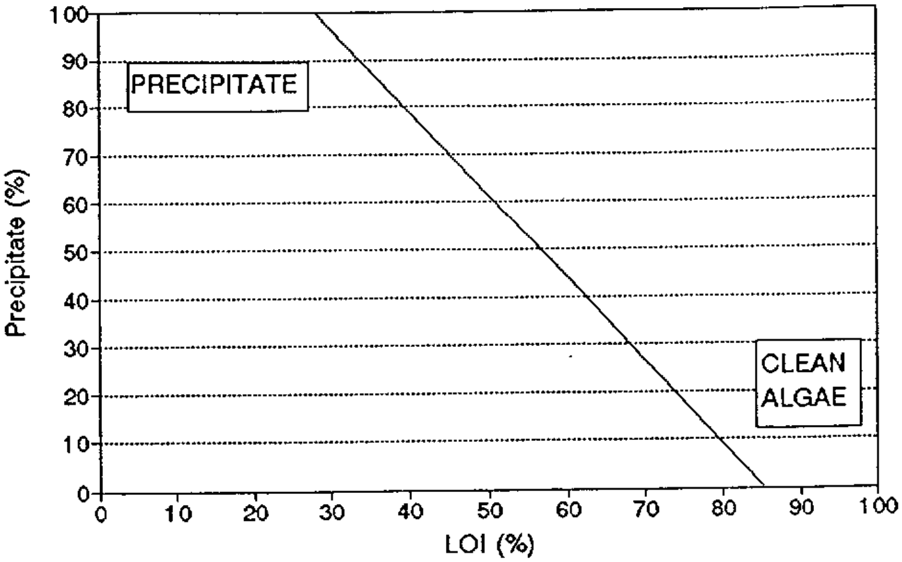


FIG 4: PPC ELEMENTAL ANALYSES
Oriental East Outflow Path

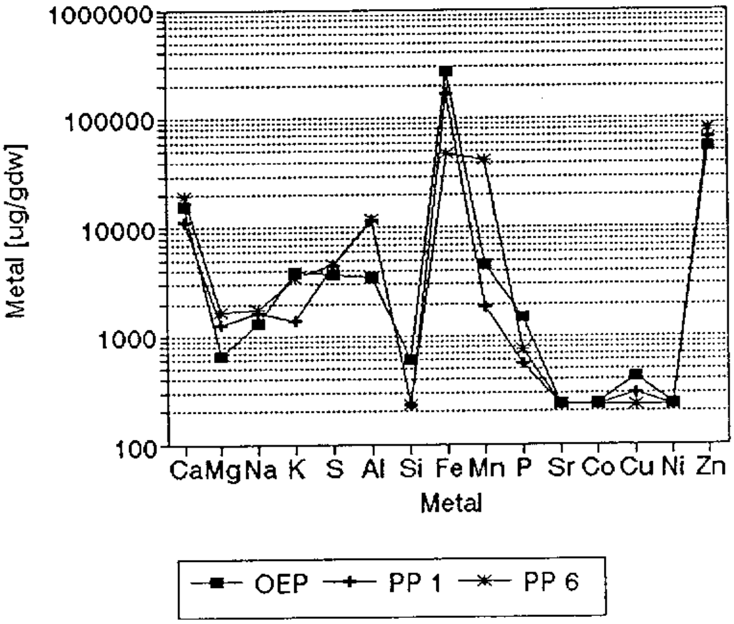


FIG 5: BRANCH PPC BIO/MASS
Polishing Ponds

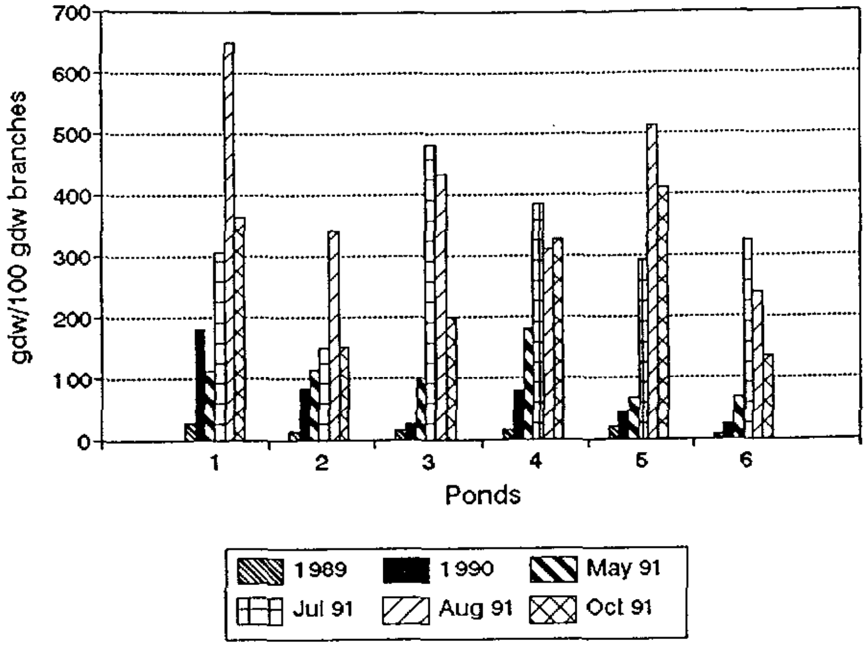


FIG 6: PPC BIO/MASS ACCUMULATION RATES
Polishing Ponds

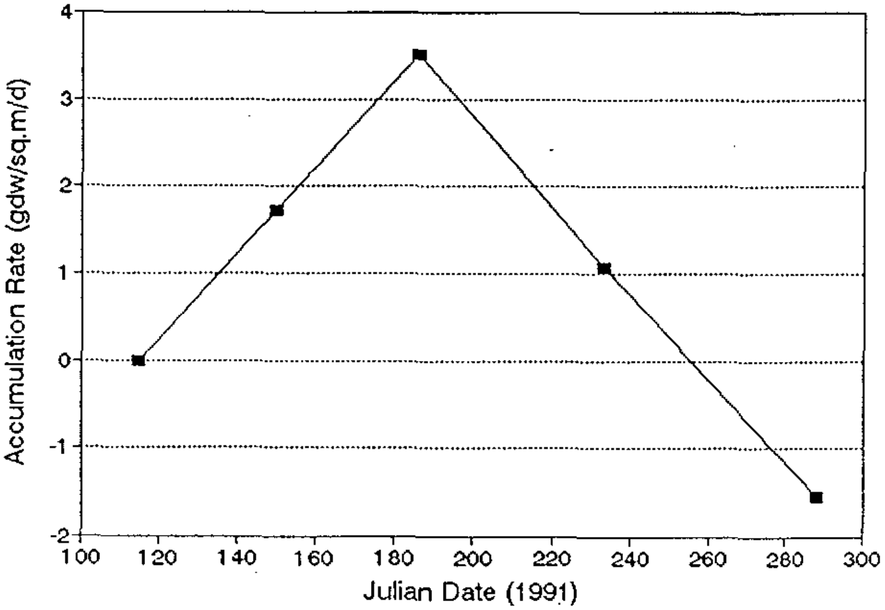


FIG 7: PERITRAP ACCUMULATIONS
Net + Substrate + Bag

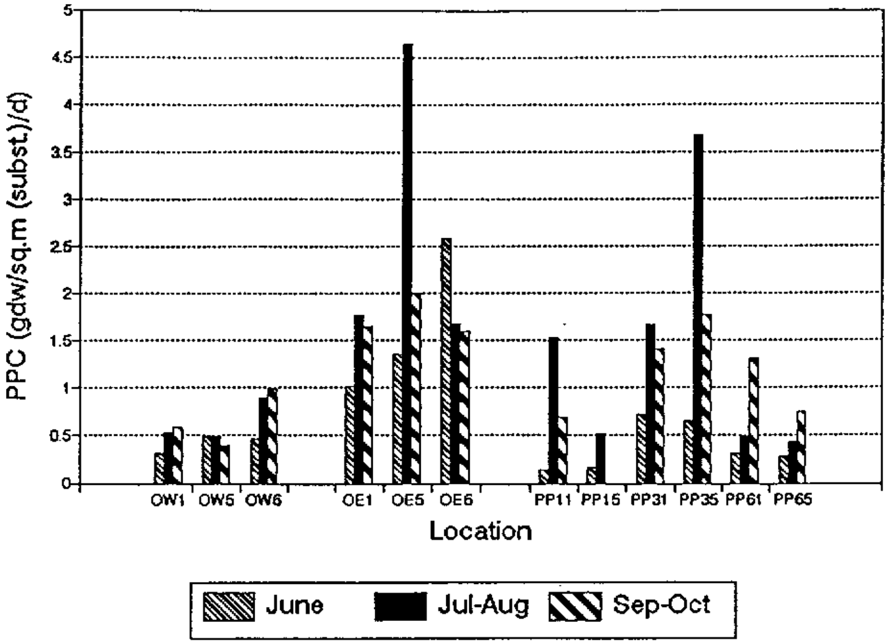


FIG 8: PPC TRAP vs. BAG WEIGHTS
Polishing Ponds

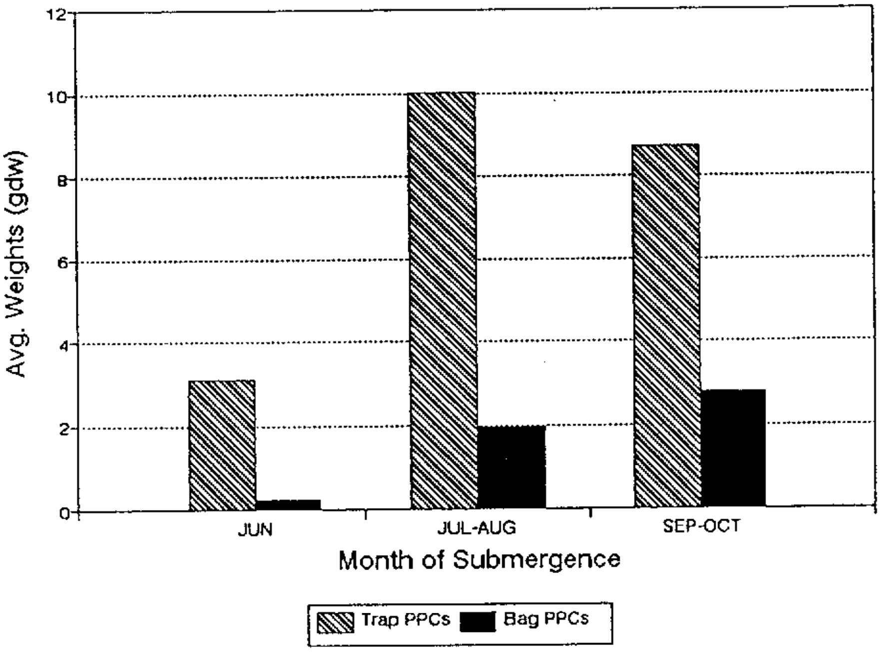


FIG 9: NEW vs. OLD BRANCHES
PPC Colonization

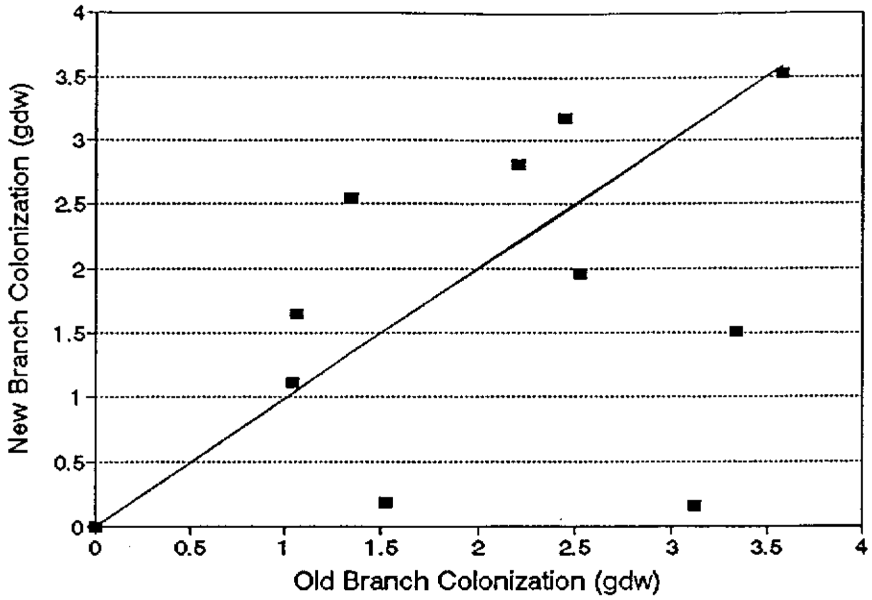


FIG 10: PERIPHYTON GROWTH RATES
Gloryholes and Polishing Ponds

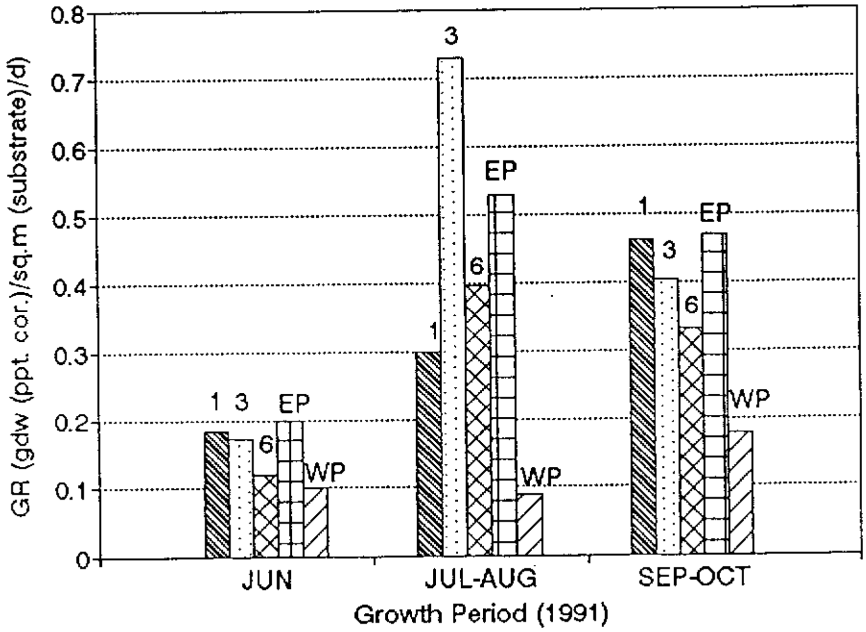


FIG 11: PERIPHYTON GROWTH RATES
Netting Growth - South Bay/Buchans

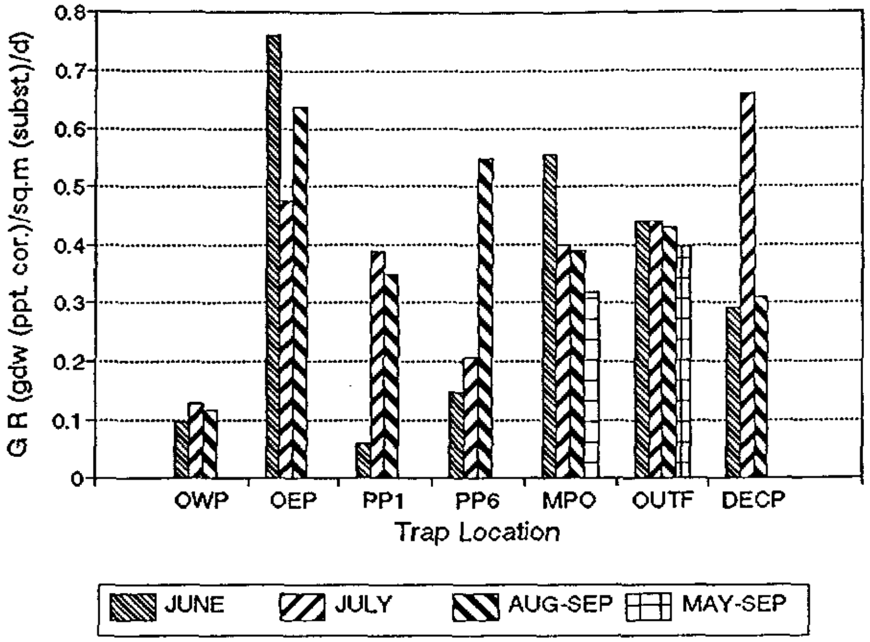


FIG 12: PPC PHOSPHORUS CONCENTRATIONS
Averages

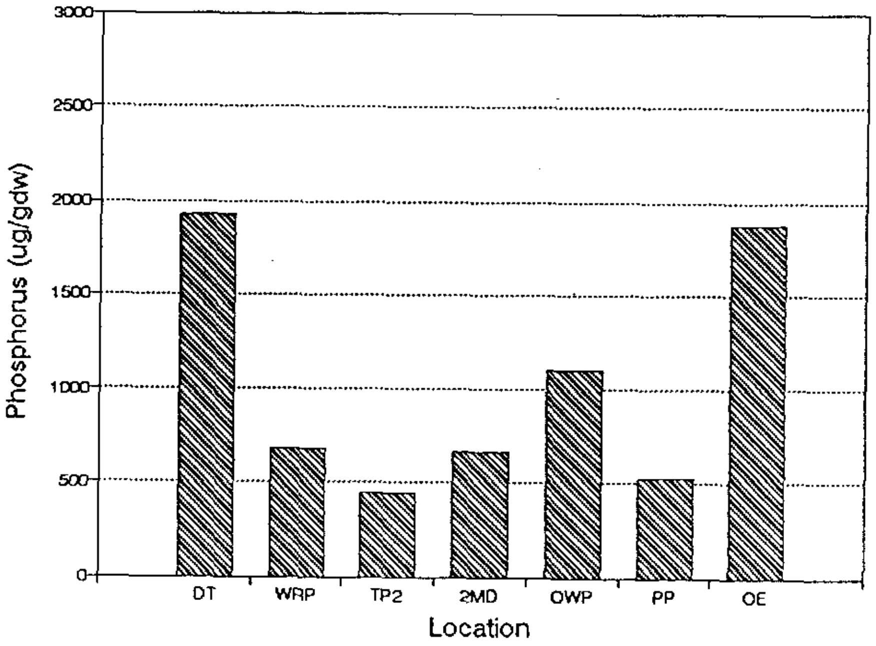


FIG 13: PHOSPHORUS CONCENTRATIONS
Fertilized vs. Unfertilized Populations

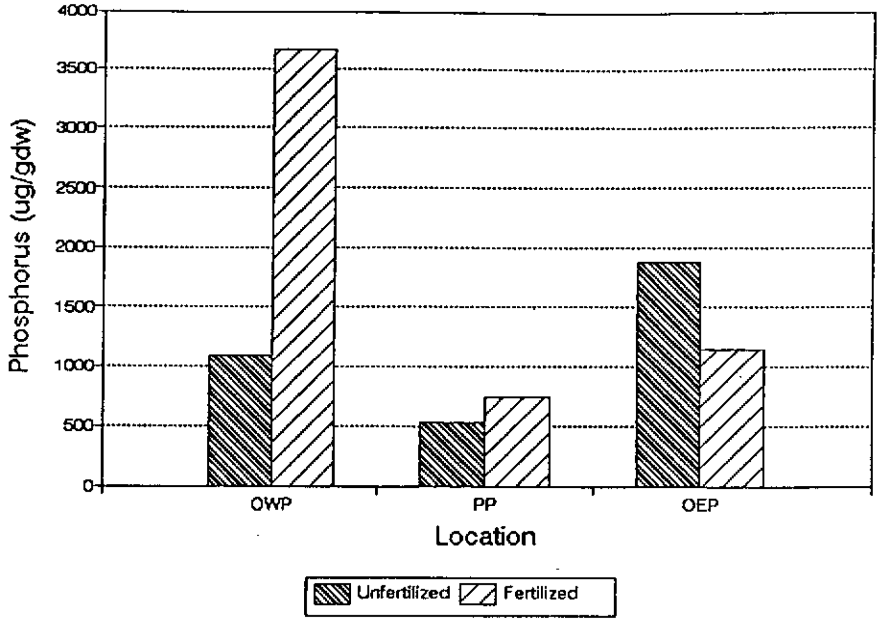


FIG 14: PPC ZINC CONCENTRATIONS
Precipitate Content vs. Zinc

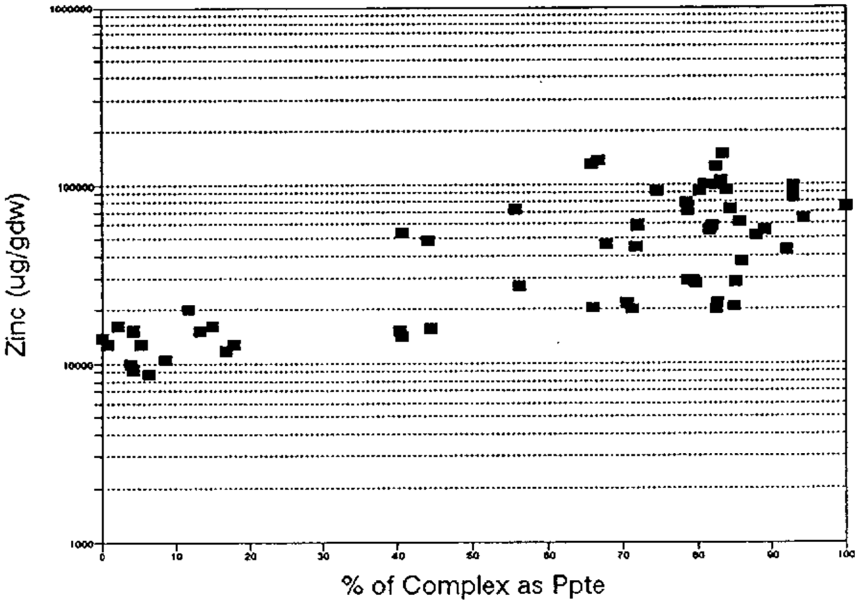


FIG 15: PPC ZINC CONCENTRATIONS

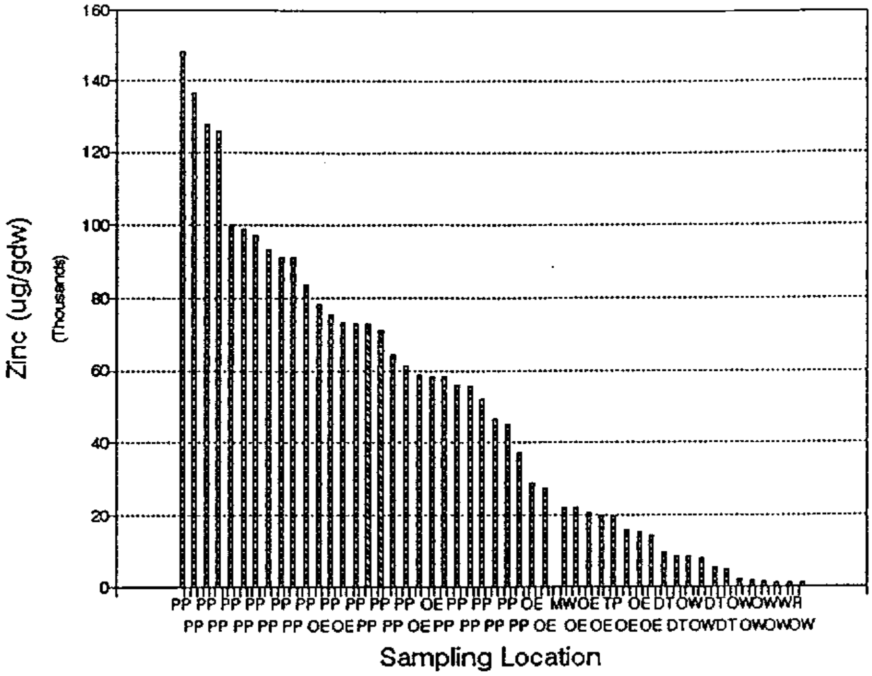


FIG 16: PPC ZINC CONCENTRATIONS
Oriental East Gloryhole Outfall

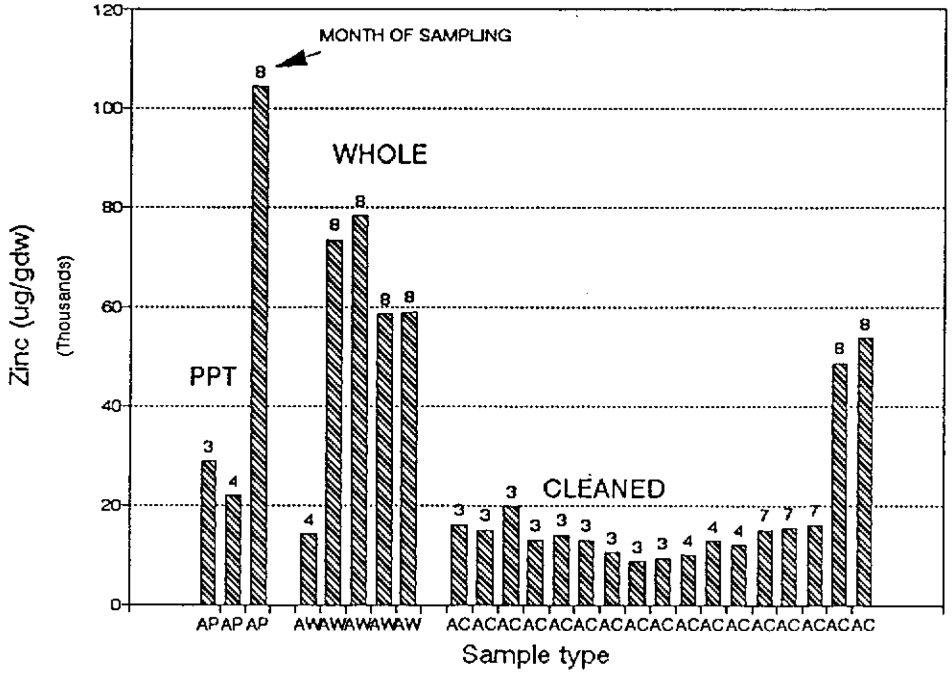


FIG 17: ZN UPTAKE EXPERIMENT
August 1991

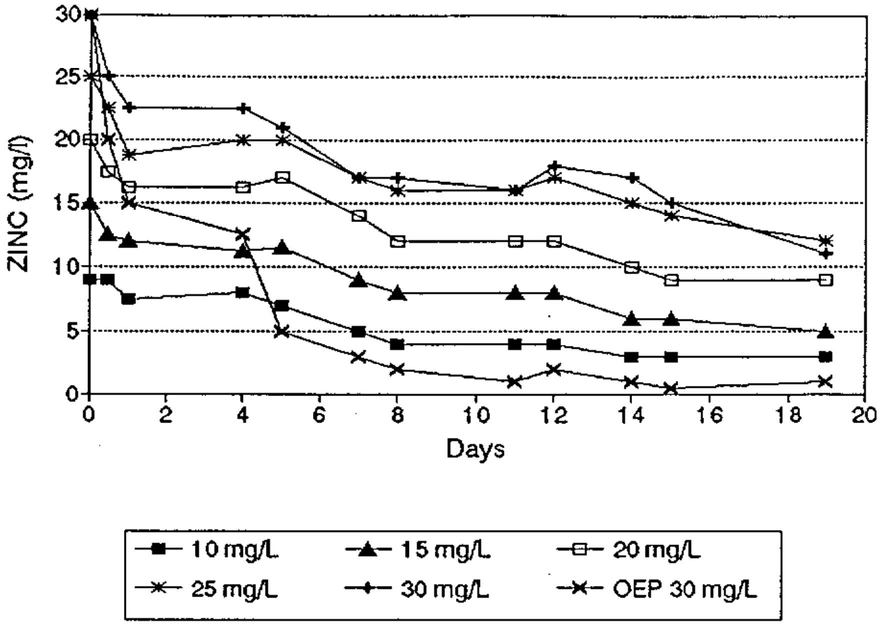


FIG 18: ZINC CONCENTRATION WATER/ALGAE
Based on Uptake of Zinc from Medium

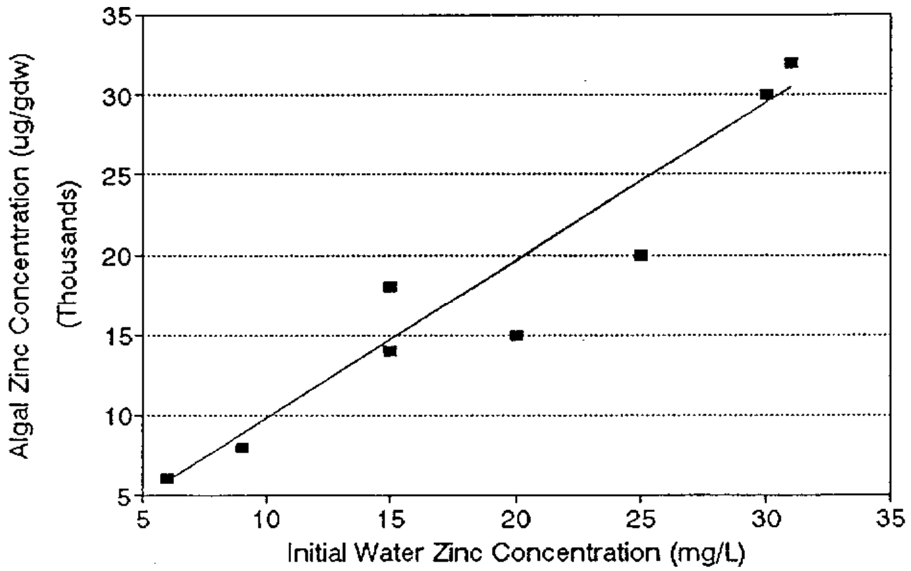


FIG 19: ZINC REMOVAL RATES
Polishing Ponds 1, 3, 6

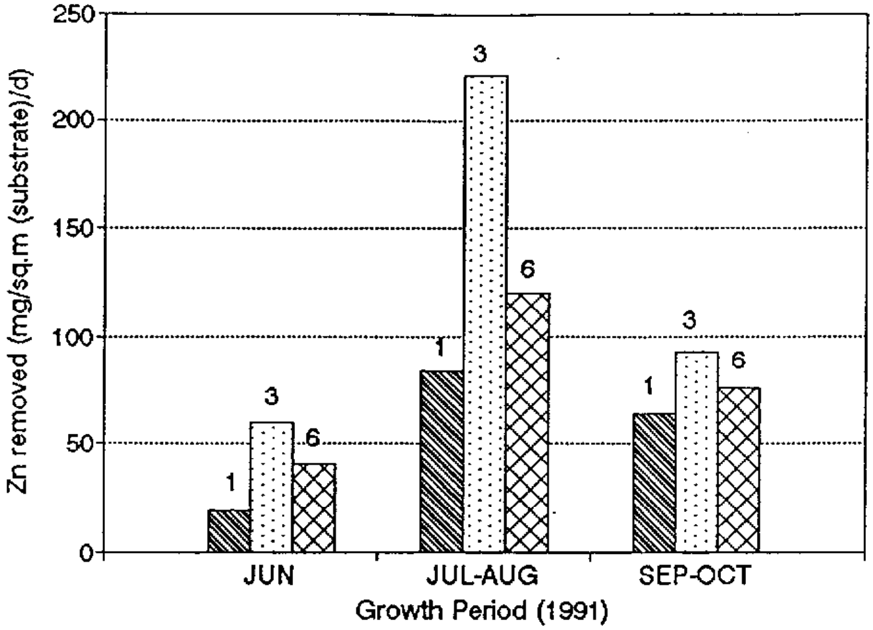


FIG 20: PPC GROWTH RATES vs. ZINC

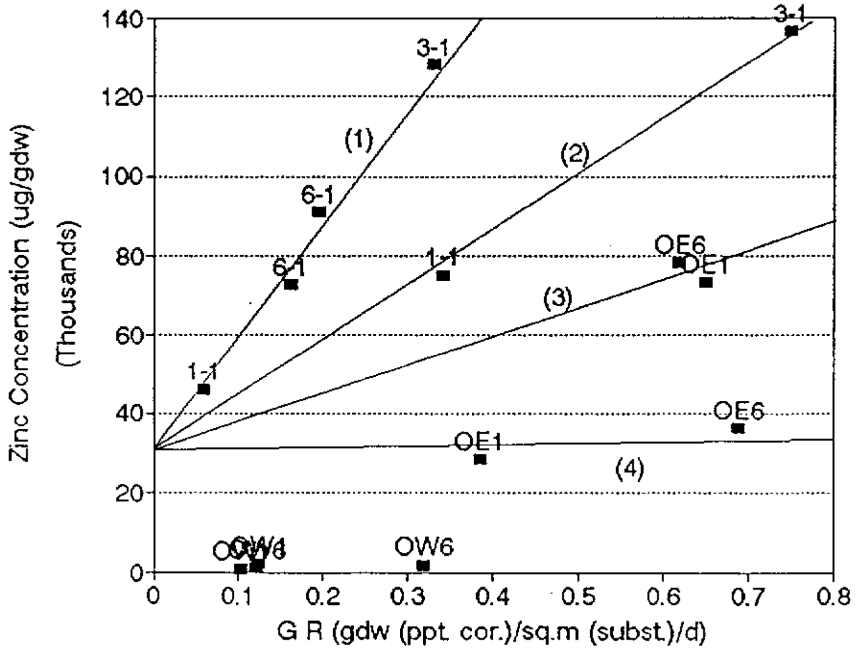


FIG 21: ZINC REMOVAL - MASS BALANCE
Loading vs. PPC Removal, August 1991

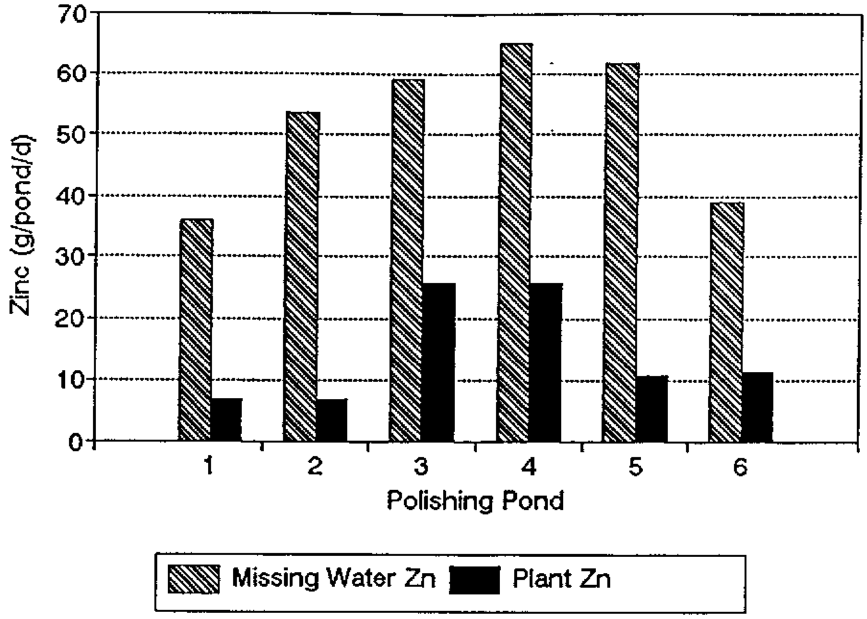


FIG 22: ESTIMATED SOLAR IRRADIANCE
Central Newfoundland

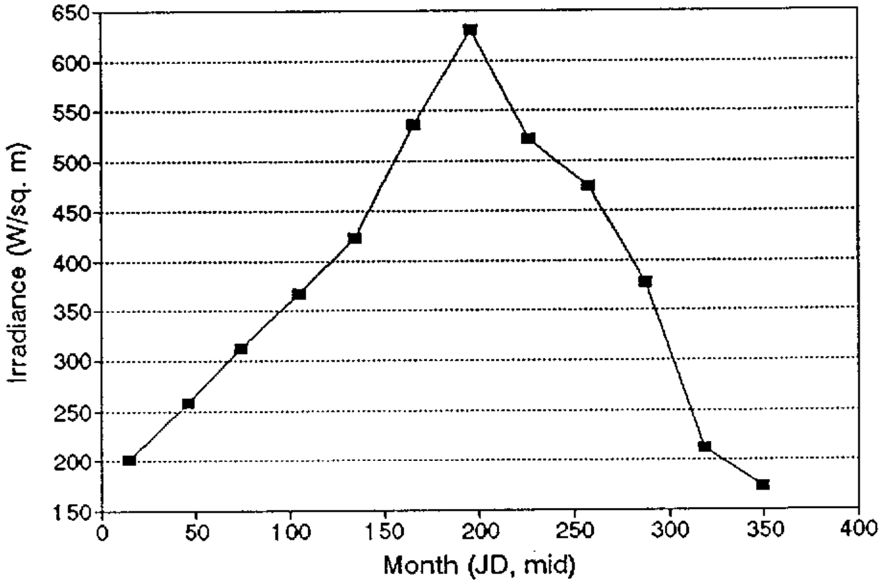
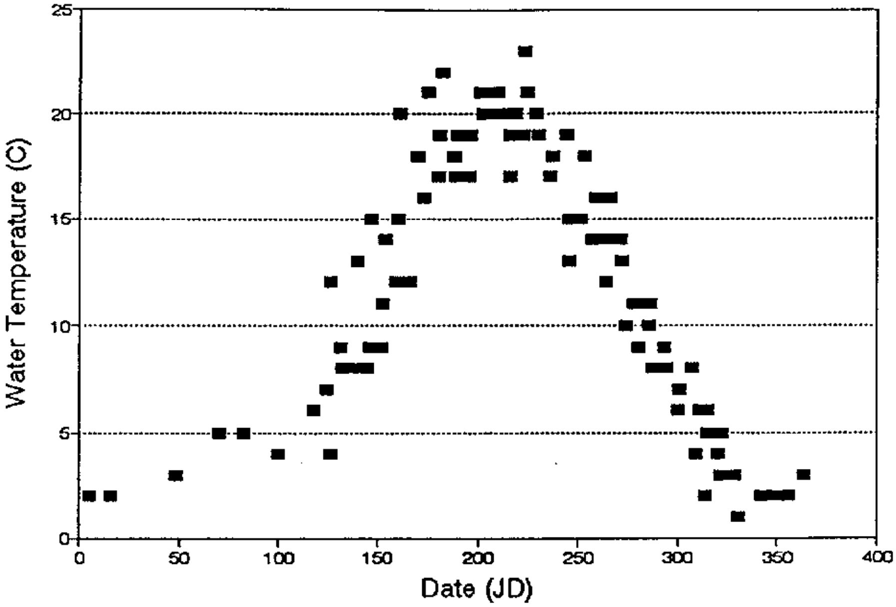
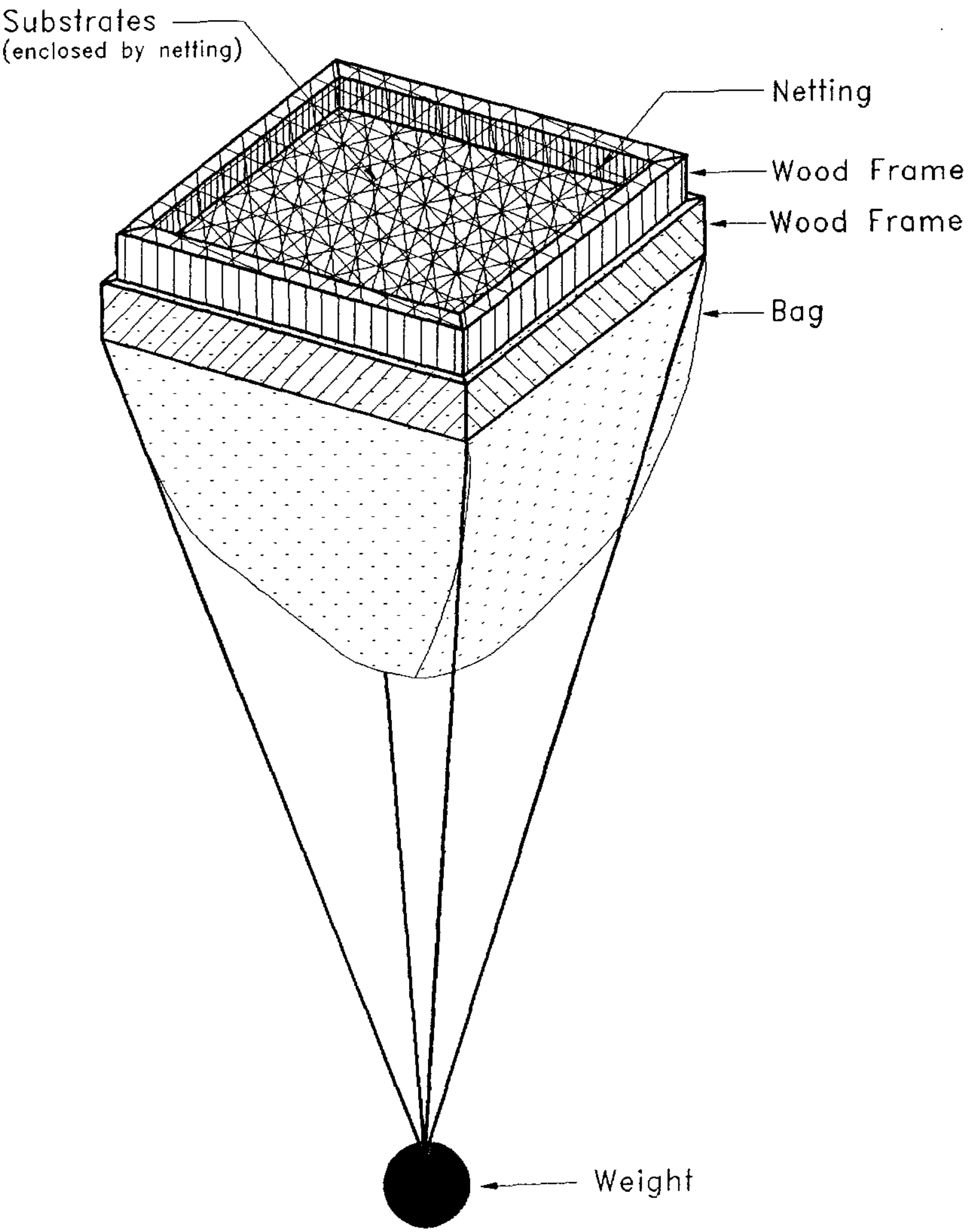


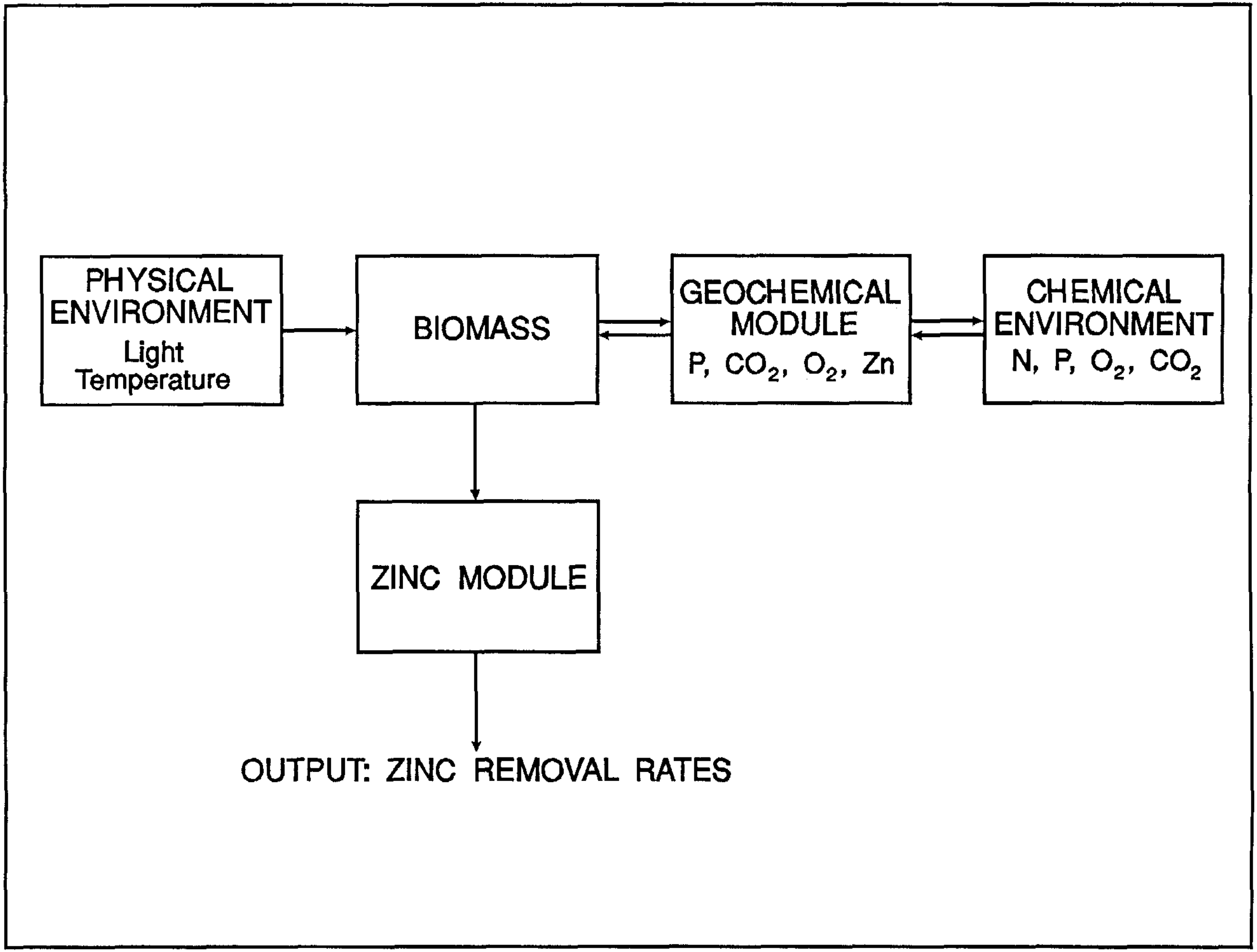
FIG 23: SEASONAL WATER TEMPERATURE
OEP OUTFLOW - 1987-1991



SCHEMATIC OF PERITRAP

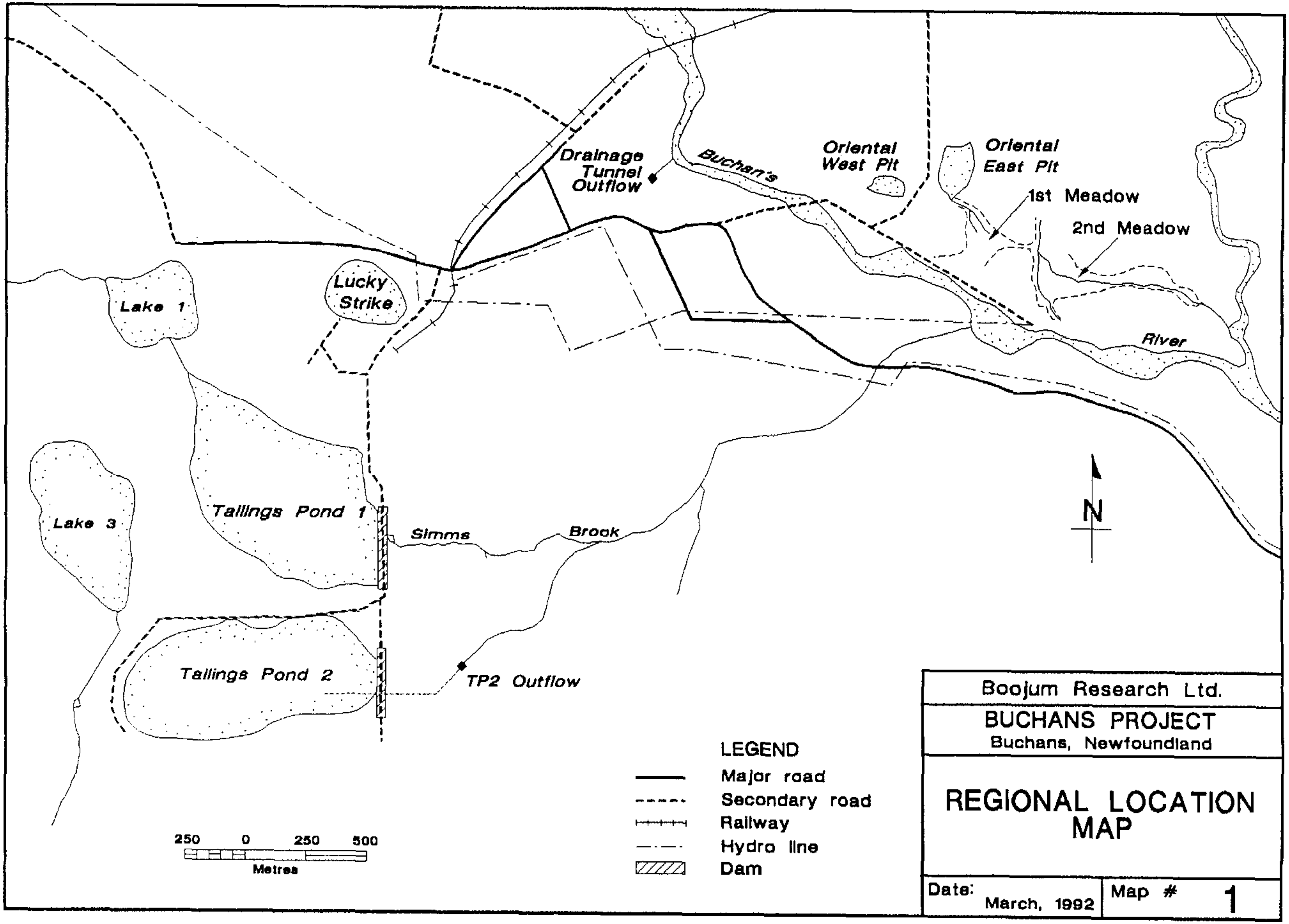


SCHEMATIC 1



PHYSICAL ENVIRONMENT
Light
Temperature
BIOMASS
GEOCHEMICAL MODULE
P, CO_2, O_2, Zn
CHEMICAL ENVIRONMENT
N, P, O_2, CO_2
ZINC MODULE
OUTPUT: ZINC REMOVAL RATES

SCHEMATIC 2



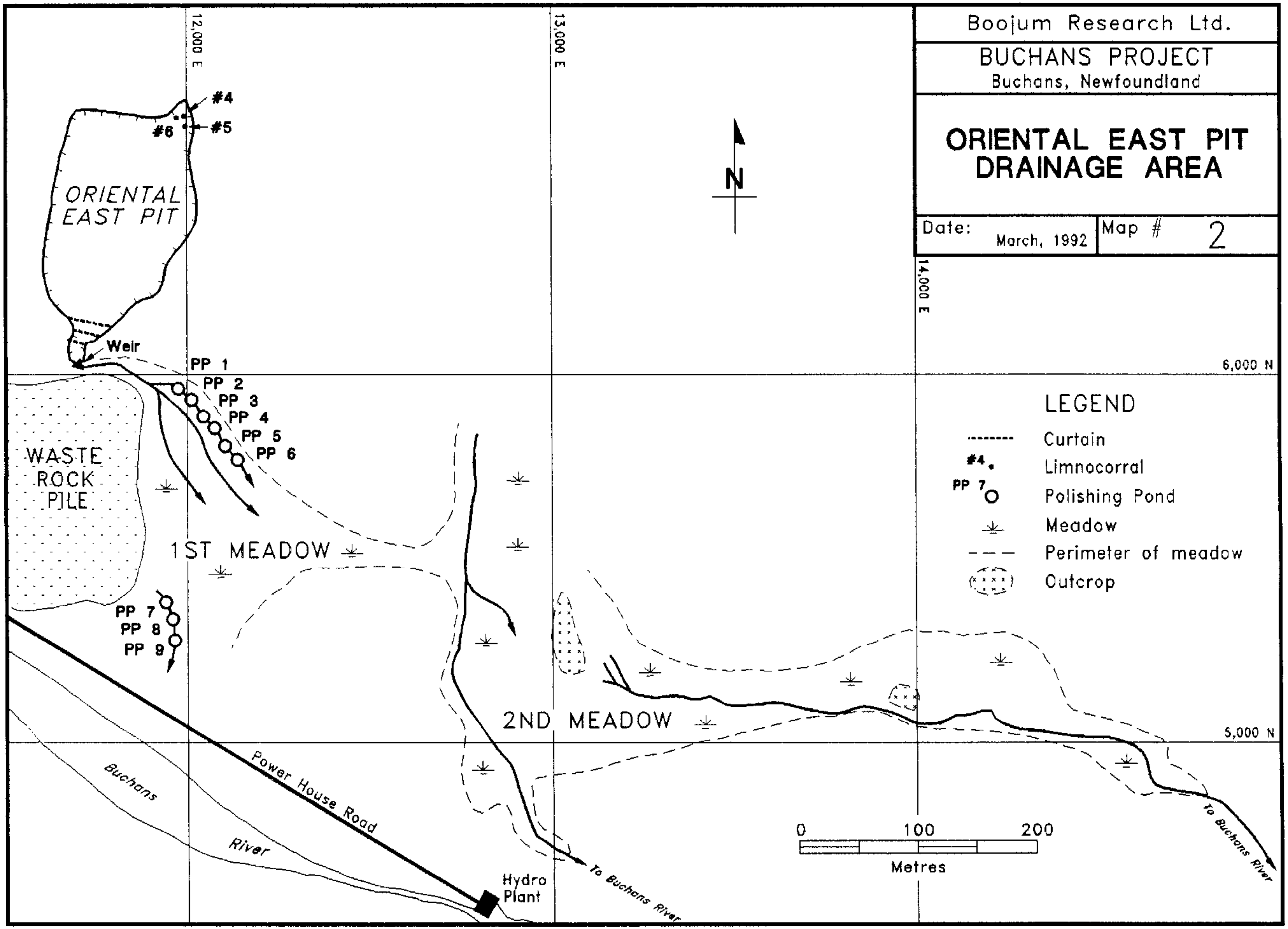
Boojum Research Ltd.

BUCHANS PROJECT
Buchans, Newfoundland

REGIONAL LOCATION MAP

Date: March, 1992

Map # 1



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BUCHANS PROJECT
Buchans, Newfoundland

ORIENTAL EAST PIT DRAINAGE AREA

Date: March, 1992

Map # 2

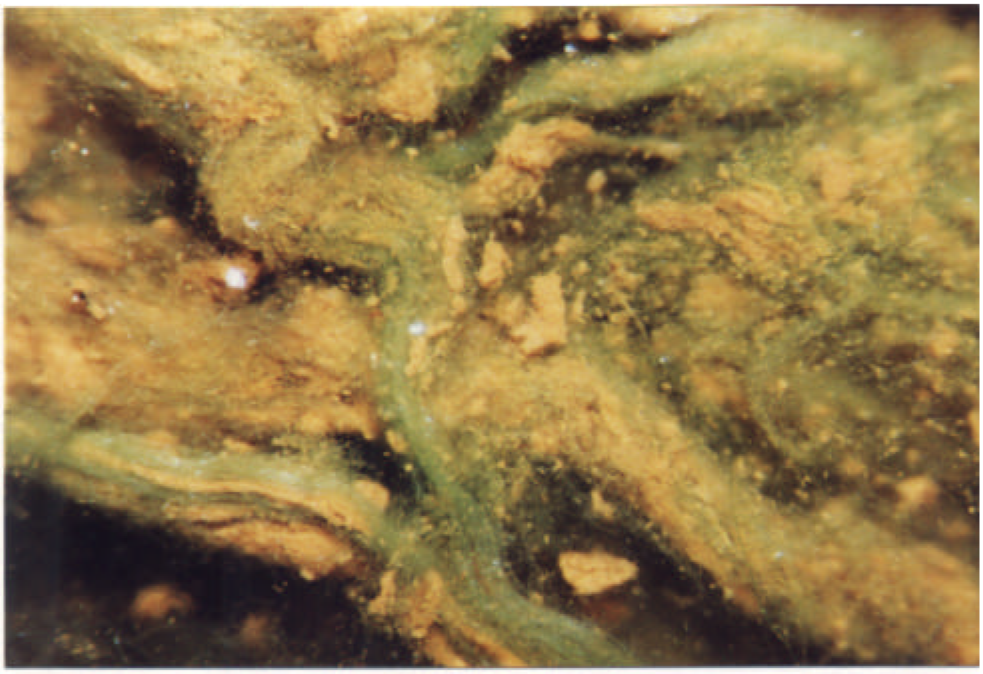

Plate 1: Microphotograph of PPC's showing periphyton/precipitate

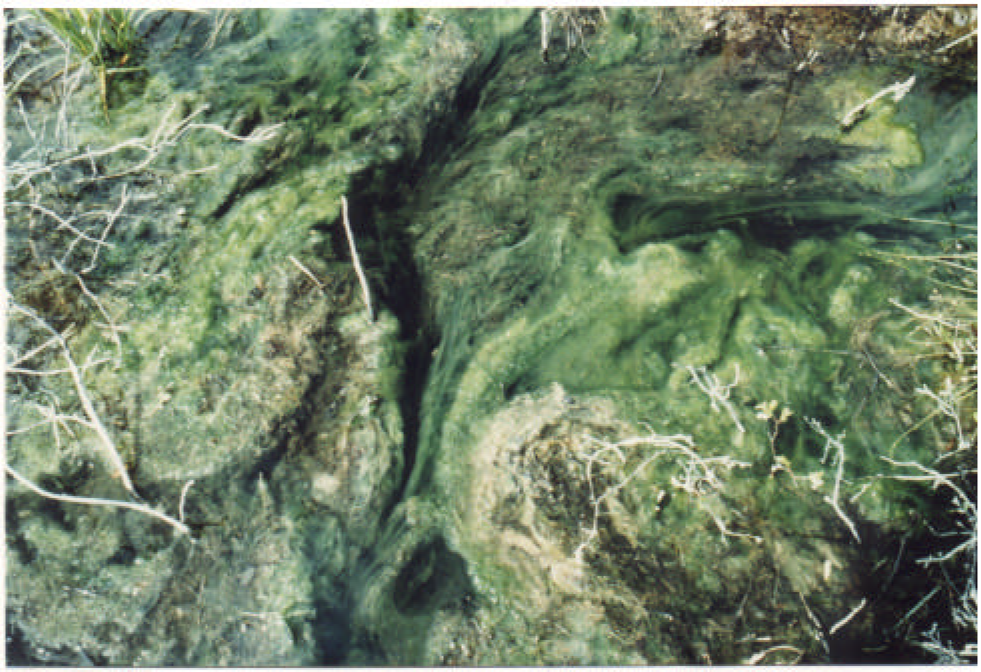

Plate 2: Photograph of "clean" seepage periphyton from 2nd meadow