

**THE EFFECTS OF ONE YEAR OF FERTILIZATION ON THE PRIMARY  
PRODUCTIVITY OF THE ARROW RESERVOIR**

by

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## ABSTRACT

The Arrow Reservoir, which consists of an upper and lower basin separated by a narrows region, was created with the impoundment of the Upper and Lower Arrow Lakes in 1969. This, followed by the construction of the Mica (1974) and Revelstoke (1983) dams upstream on the Columbia River, the major tributary to the reservoir, has had a significant impact on the aquatic ecosystem. Preliminary studies conducted in 1997 and 1998 revealed that the nutrient load to the reservoir was likely insufficient to support the ecosystem. A fertilization and monitoring project was initiated in 1999 with the intent of reversing the oligotrophication of the reservoir.

The Arrow Reservoir was studied in 1998, the pre-treatment year, and in 1999, the first year of fertilization, to examine the effects of one year of fertilization on the activity and abundance of bacteria and phytoplankton. Primary productivity and phytoplanktonic abundance were estimated from radiolabelled bicarbonate incorporation studies and chlorophyll-a analyses, respectively. Bacterial activity was examined using  $^{14}\text{C}$ -glucose incorporation studies along with CTC staining and counting, while DAPI staining and counting was employed for bacterial abundance estimates.

Samples were collected on a monthly basis from May to September in 1998 and from April to September in 1999. Six depths (the surface, 1, 2, 5, 10 and 15 m) were sampled at a single, central location in each basin. In addition to these vertical transects, horizontal transects of the upper basin were performed in June, August and September of 1999. Horizontal transect samples were collected at a depth of 2 m at 6 approximately equidistant locations in a north-south transect of the basin.

The primary productivity and bacterial activity and abundance in the upper basin, where nutrient addition occurred, appear to have been enhanced by fertilization, as these parameters were each significantly greater in this basin in 1999 than in 1998. The lower basin did not appear to benefit from the nutrient enrichment during the first year of fertilization as these parameters did not differ significantly in this basin from 1998 to

1999. A decrease in algal biomass in the lower basin from 1998 to 1999, which was likely a function of the colder, wetter year in 1999, was not observed in the fertilized upper basin.

Algal bioassays using *Selenastrum capricornutum* were used to assess the bioavailability of nutrients in the tributaries of the Arrow Reservoir. The nitrogen and phosphorus concentrations of all of the tributaries assayed were severely growth limiting. The majority of tributaries were nitrogen and phosphorus co-limiting, while samples from the agriculturally-influenced Innonoaklin Creek were limited to a greater extent by nitrogen.

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## 1. INTRODUCTION

### 1.1 General Introduction

The effects of the construction and operation of a dam on the surrounding aquatic ecosystem are numerous and can be profound. The nature and severity of these effects are different in each situation as many variables are involved. The most obvious impact on the ecosystem is the loss of habitat due to the physical barrier preventing migration and the flooding above the dam.

Several less obvious effects may be caused by the altered flow of water, sediments and nutrients. Altered flow regimes and limited sediment throughflow may alter habitat, predator prey interactions and life cycles of the flora and fauna (Power et al. 1996; Ligon et al. 1995).

The retention of nutrients within a reservoir is often at the expense of downstream lakes and reservoirs. These downstream waters suffer from declining nutrient concentrations, a process known as oligotrophication. The impacts of oligotrophication are felt at all trophic levels within an ecosystem with the possible collapse of fish populations (Ney 1996). A more thorough discussion of the impacts of reservoirs on aquatic ecosystems is provided in section 2.1.4.

The Arrow Lakes, part of the Upper Columbia River in the Kootenay region of southwestern British Columbia, were impounded in 1969. This, followed by the construction of two upstream dams, Mica (1974) and Revelstoke (1983), has had considerable impact. Following construction of the dams, significant habitat losses occurred due to flooding and inaccessibility (Lindsay and Seaton 1978, Martin 1976). In addition, the nutrient concentrations in this reservoir have declined as a result of upstream impoundments.

The limnology and ecology of the Arrow Reservoir were studied during the period of 1997 to 1999 (Pieters et al. 1998). The results of these studies indicate that the nutrient loading to the reservoir was not sufficient to support the ecosystem. The potential for the collapse of first planktivorous fish, followed by large piscivorous fish was considered to be significant.

A five-year experimental fertilization program, funded by the Columbia Basin Fish and Wildlife Compensation Program (CBFWCP) was initiated in the Arrow Reservoir in April 1999. As a part of this program, the primary productivity of the reservoir was monitored prior to fertilization from May to September 1998 and during fertilization from April to September 1999.

## 1.2 Arrow Reservoir General Information

The Arrow Reservoir is a long, narrow, fjord-like body of water situated between the Selkirk and Monashee mountain ranges in the southern interior of British Columbia, Canada (Figure 1.1). Prior to impoundment the Lower and Upper Arrow Lakes were separated by a river. Currently, a river-like region, referred to as the Narrows, separates the two basins only during periods of low flow. The maximum depths of the upper and lower basins of the Arrow Reservoir are approximately 300 and 200 m, respectively. The combined surface area of upper basin and the narrows region at full pool is 29,557 ha, while that of lower basin is 16,893 ha (Pieters et al. 1998).

The pre-impoundment maximum and mean water level fluctuations in the Arrow Lakes were 11.4 m and 8.0 m, respectively. Following impoundment, the maximum water level variation is 19.6 m and the mean variation is 15.5 m. Prior to impoundment, water levels remained consistently low throughout the year except during spring freshet. In contrast, since the construction of the Keenleyside dam, water levels remain high during most of the year with the level dropping significantly during the winter (Pieters et al. 1998).

The population surrounding the Arrow Reservoir is relatively small. The estimated 1999 populations of the communities of Revelstoke and Nakusp, which are located within the Arrow Reservoir drainage basin, are 8226 and 1788, respectively (Government of British Columbia Ministry of Finance and Corporate Relations web site: <http://www.bcstats.gov.bc.ca/data/pop/pop/mun9699e.htm>). The few additional communities in this drainage basin are significantly smaller.

Prior to impoundment, the Arrow Lakes supported populations of numerous fish species, including the following sport fish: rainbow trout (*Oncorhynchus mykiss*), bull trout (*Salvelinus confluentus*), kokanee (*O. nerka*), white sturgeon (*Acipenser*

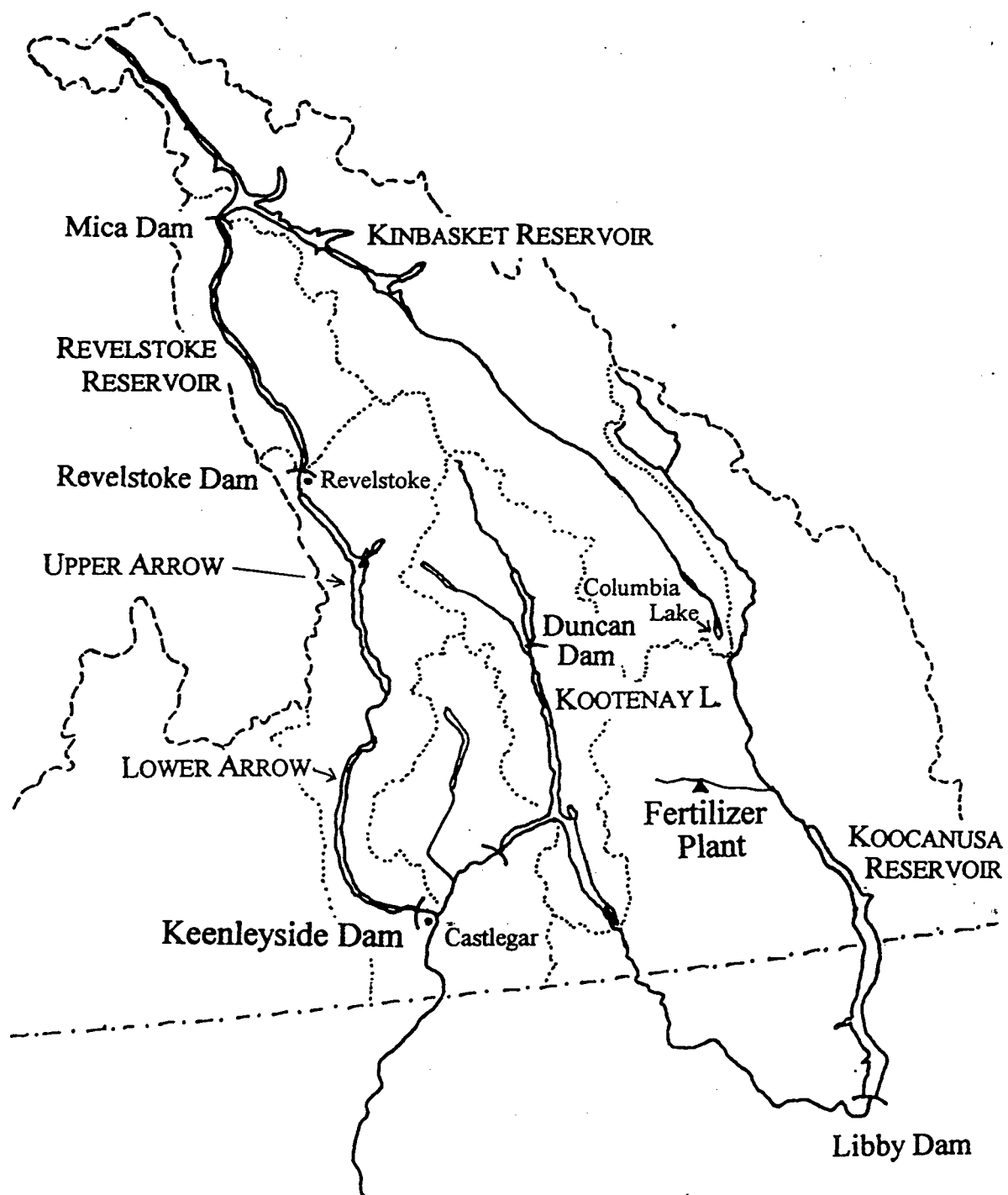


Figure 1.1 – Map of the Canadian portion of the Columbia River Basin

*transmontanus*), mountain whitefish (*Prosopium williamsoni*) and burbot (*Lota lota*) (Pieters et al. 1998). However, prior to the start of the lake fertilization project it was feared that the nutrient load to the lake was not sufficient to continue supporting these populations.

### **1.3 Project Scope and Objectives**

The primary objective of the fertilization project is to reverse the oligotrophication of Arrow Reservoir and enhance the dwindling fish populations. The main target species are kokanee, rainbow trout and bull trout.

The main objective of this thesis project is to determine whether one season of fertilization of Upper Arrow Lake has any effect on the primary production of the Arrow Reservoir. A sample program was carried out in the Arrow Reservoir throughout the growing season during the year prior to fertilization (May to September 1998) and again during the first year of fertilization (April to September 1999). An additional study was performed to assess the bioavailability of nutrients in the tributaries and to determine which nutrient(s) is(are) limiting.

## 2. LITERATURE REVIEW

### 2.1 General limnology/lake ecology

This section provides a general discussion of the pertinent biotic and abiotic features of lakes. A brief description of the important characteristics of the various groups of organisms most relevant to this thesis is also given.

#### 2.1.1 Abiotic Features

##### *Relevant physical features*

Within a lake, the physical, chemical and biological characteristics of the water and the substrate are not uniform. As sunlight penetrates the water, some is transmitted, while the remainder is reflected, refracted or absorbed. The adsorption of light is essentially the transformation of light energy into heat.

Since only a portion of the sunlight is transmitted, the intensity decreases logarithmically with depth (Wetzel 1983, Horne and Goldman 1994). This has two profound impacts on the structure of the lake. The absorption of light in the surface waters results in a thermal stratification of the water (Wetzel 1983). The transition zone between the two layers, where the change in water temperature with depth is the greatest is called the metalimnion. The warm surface layer is referred to as the epilimnion, while the colder water below the metalimnion is called the hypolimnion. In addition, since primary producers require light energy, they are restricted to the water where the sunlight intensity is sufficient to support photosynthesis. The extent to which light is transmitted through water depends on the quantity of dissolved and suspended material in the water.

Since light is absorbed by water, the light intensity will be just sufficient for photosynthesis to be equal to respiration over a 24 hour period (i.e. the net oxygen production is zero) at a certain depth, the compensation depth (Lee 1989). Below this depth is the aphotic zone and above it is the euphotic zone. The compensation depth occurs where the light intensity is approximately 1% of that at the surface (Horne and Goldman 1994). Since photosynthesis can not occur below the compensation depth, primary production occurs primarily in the euphotic zone.

A lake may be divided into several regions based in part on the compensation depth (Lee 1989). The bottom of the lake is referred to as the benthic zone. The intersection of the compensation depth with the benthic zone forms the lower border for a region which includes the littoral and sublittoral zones. The littoral zone is the shallow region of the lake perimeter where the light intensity is sufficient to support periphyton (attached algae) and rooted macrophytes. The region bordered at the top by the maximum depth of rooted vegetation and at the bottom by the intersection of the compensation depth with the lake bottom is the sublittoral zone. The region outside the littoral and sublittoral zones and above the compensation depth is the pelagic zone while the region below the compensation depth and above the benthic zone is the profundal zone. In addition to their physical distinctions, these regions are biologically distinct and organisms may be classified based on the zone in which they live.

#### *Relevant chemical features of lakes*

Along with the climate, the size, topography, geology and vegetation of the watershed are important factors determining the chemical composition of the lake (Horne and Goldman 1994). These factors influence the rate of erosion and dictate the physical and chemical properties of the eroded material.

The atmosphere is also a source of nutrients for a lake. Some gaseous compounds, such as carbon dioxide, ammonia and nitrogen, dissolve directly into the water. In addition, many chemicals are delivered to a lake in an air-borne particulate form or in rainwater. Sodium, chlorine, bromine, iodine and fluorine present in moisture evaporated from the ocean is delivered to the terrestrial environment through precipitation (Horne and Goldman 1994).

In addition to these natural processes, human activities may severely impact aquatic chemistry. Deforestation leads to increased erosion, delivering greater quantities of sediments and nutrients to a lake. Agricultural practices, such as fertilization and weed and pest control, may result in an input of nutrients, herbicides and pesticides into the aquatic environment.

Within a water body, various nutrients undergo physical, biological and chemical transformations. The phosphorus and nitrogen cycles within a lake are well understood and have been described in detail in Horne and Goldman (1994) and Wetzel (1983). It is



worth noting some of the important nutrient transformations which occur within a lake. Dissolved nutrients become part of the particulate pool when they are absorbed by organisms and incorporated into cellular material. Excretions from or decomposition of these organisms returns these nutrients to the soluble pool. Nutrients which are part of the particulate pool may settle out of the water column and become part of the sediment in the benthic zone. Phosphate has a relatively high affinity for particulate matter and will reversibly adsorb to suspended particles. Phosphate may also precipitate with ions such as  $\text{Ca}^{2+}$  and  $\text{Fe}^{2+}$ .

Under anoxic conditions, which often occur adjacent to and within the sediments, soluble phosphate is released from particulate matter and biological nitrogen fixation and denitrification may occur. Phosphate releases from anoxic sediments may be up to 1000 times faster than those from oxygenated sediments (Horne and Goldman 1994). Under well oxygenated conditions, bacteria may oxidize ammonia to nitrate, a process known as nitrification. It is also important to note that phosphorus, lacking a gas phase, can not be replenished by reductive assimilation (photosynthesis) as nitrogen and carbon can.

Of the elements required to support life, phosphorus is often the one with the lowest availability to requirement ratio and is therefore often the limiting nutrient. However, in some circumstances phosphorus is relatively abundant due to pollution or erosion of phosphorus rich soils in a watershed. In these cases nitrogen is usually the limiting nutrient. According to Horne and Goldman (1994), phosphorus is considered to be limiting when the nitrogen to phosphorus ratio (N:P) by weight is greater than 10, while nitrogen is limiting when this ratio is less than 10.

Lakes may be classified based upon their nutrient content. In general, lakes that are rich in nutrients are classified as eutrophic, while nutrient deficient lakes are defined as oligotrophic. The classification of the lake is often assigned based on the phosphorus concentration in the lake. A lake is considered to be oligotrophic if the total phosphorus concentration is in the range of 5-10  $\mu\text{g/L}$  and eutrophic in the range of 30-100  $\mu\text{g/L}$ . At 10-30  $\mu\text{g/L}$  a lake is classified as mesotrophic and at less than 5  $\mu\text{g/L}$  or greater than 100  $\mu\text{g/L}$  the lake is ultraoligotrophic or hypereutrophic, respectively (Bronmark and Hansson 1998).

### 2.1.2 Biotic features of lakes

As previously mentioned, the organisms in a lake may be described or classified based on the region which they inhabit. This thesis deals exclusively with organisms which live in the pelagic region. Pelagic organisms which are strong swimmers and move freely throughout the water are referred to as nekton, while those that are weak swimmers or non-motile are called plankton. The autotrophic phytoplankton and heterotrophic bacterioplankton and zooplankton are members of the plankton community.

Some organisms occupy different habitats during different life stages. For example, kokanee, which are pelagic fish as adults, usually lay their eggs in tributaries to the lake where they spent their adult life (Ford et al. 1995). After hatching, the young migrate to the lake where they inhabit the littoral region until they are large enough to survive in the pelagic region.

There is a large variety of organisms which inhabit aquatic ecosystems. They each have different biotic and abiotic requirements. Therefore, the species present in a given ecosystem will depend upon the physical, chemical and biological features of that ecosystem.

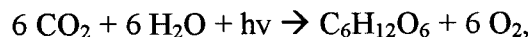
The following sections describe in detail the important characteristics of the aquatic organisms most relevant to this thesis. In addition, the interactions between these organisms are briefly discussed.

#### *Bacteria*

Bacteria play a very important role in the aquatic ecosystem as they are one of the main groups of organisms involved in mineralization. By degrading dissolved organics and incorporating them into cellular material they effectively recycle some of the nutrients which would otherwise be lost from the system (Horne and Goldman 1994). In addition some bacteria perform such vital tasks as nitrogen fixation, nitrification and denitrification, as well as sulfur oxidation and photosynthesis. They are ubiquitous and form a relatively abundant food source for protozoans (heterotrophic flagellates and ciliates) and other larger zooplankton.

### *Primary producers*

All primary producers utilize energy from the sun to convert carbon dioxide and water into energy-rich glucose. This process is known as photosynthesis and may be represented as follows:



where  $h\nu$  is light energy. It is worth noting that oxygen ( $\text{O}_2$ ), a vital molecule for aquatic life, is produced by photosynthesis.

All photosynthetic organisms possess the pigment chlorophyll a, which captures sunlight energy and converts it into chemical energy. The absorption maxima for this pigment are at 430 and 660 nm (Bronmark and Hansson 1998). Some primary producers also possess one or more accessory pigments whose absorption maxima differ from that of chlorophyll a. These accessory pigments, such as chlorophylls b and c, carotenoids and biliproteins, pass the captured energy on to chlorophyll a (Wetzel 1983). This allows the primary producer to use a wider range of wavelengths, an important attribute considering the changing characteristics of light with depth.

Primary producers may be divided into two broad categories: the macrophytes and algae. Each of these categories contain free-floating and substrate-associated species.

### *Macrophytes*

While free-floating macrophytes obtain their nutrients exclusively from the water, substrate-associated forms may extract nutrients from the sediments through their roots. Different forms of macrophytes tend to thrive at different depths forming a zone of transition within the littoral zone. The pattern of zonation, starting from the shoreline, generally begins with emergent macrophytes, followed by floating-leaved forms and then submersed forms. (Bronmark and Hansson 1998, Horne and Goldman 1994)

Macrophytes play several important roles in the aquatic ecosystem. By removing nutrients from the sediments, rooted forms recycle these nutrients back into the aquatic environment. Macrophytes provide vital habitat for aquatic invertebrates and small fish. They are also a food source for a variety of animals, either directly or through the film of algae and microscopic animals that build up on the surface of the plants.

### *Algae*

The term algae refers to a highly diverse group of organism. They may be divided into two main classes: the free-floating phytoplankton and the substrate associated periphytic or attached algae. In most aquatic environments, algae make a more significant contribution to primary production than macrophytes (Horne and Goldman 1994). They encompass a wide variety of sizes and shapes and may be unicellular, filamentous or colonial.

As with attached macrophytes, periphytic algae are found along shorelines or littoral regions of lakes where light penetrates to the lake bottom. If the local conditions, such as pH, current velocity and nutrient availability, are within an optimal range, periphytic algae may utilize a variety of substrates, such as macrophytes, rocks or mud.

The periphytic algae and phytoplankters have various optimal ranges of pH and nutrient availability. Therefore, the number of species present in a given lake may be a good indicator of the pH and nutrient status of that lake (Bronmark and Hansson 1998).

Some species of phytoplankton have various adaptations, such as spines, a protective mucous sheet or a large size, in order to resist grazing by their zooplankton predators (Horne and Goldman 1994). Each adaptation is most effective under specific conditions.

Phytoplankton are often classified based on their size as follows: picoplankton (0.2 – 2  $\mu\text{m}$ ), nanoplankton (2 – 20  $\mu\text{m}$ ), microplankton (20-200  $\mu\text{m}$ ) and macroplankton (>200  $\mu\text{m}$ ). The size of these organisms is important for a variety of reasons. Smaller cells have a larger surface area to volume ratio and exhibit more rapid rates of nutrient uptake. Smaller cells also sink at much slower rates as the rate of sinking is directly proportional to the effective radius (the radius of a sphere of equivalent volume for particles which are not spherical) (Horne and Goldman 1994). In addition, zooplankton predators are often limited to phytoplankton prey of specific size ranges. In this manner, many larger macrozooplankton are not capable of digesting picoplankton, while smaller nano- and microzooplankton can not utilize larger species of phytoplankton.

Due to the ever changing physico-chemical conditions of lakes, the phytoplankton composition tends to go through a seasonal succession (Horne and Goldman 1994, Wetzel 1983). As the physical, chemical and biological characteristics of a lake change,

the phytoplankton community also changes. While the species composition is characteristic of each individual lake, the seasonal succession tends to follow a general trend. There is usually one large bloom in the spring, which is dominated by various species of cold-tolerant diatoms. This spring bloom occurs following thermal stratification when plankton and nutrients are no longer mixed throughout the entire water column. Nutrients are transferred to higher trophic levels as phytoplankton are consumed and the supply of dissolved nutrients decreases. A period of low algal biomass occurs following the spring bloom due to the depleted dissolved nutrient supply in the epilimnion. Increased parasitism and zooplankton grazing in response to increased water temperatures also plays a role in the decline of the spring bloom and the subsequent period of low algal biomass. Any summer blooms are generally smaller, more irregular and dominated by flagellates and blue-green algae. As the water destratifies and the near surface water is supplied with nutrients from the hypolimnion, a fall bloom often occurs, which is generally dominated by diatoms but may also include blue green algae and dinoflagellates.

#### *Zooplankton*

Along with rotifers and two classes of crustaceans, protozoans make up the bulk of the zooplankton. Flagellate and ciliate protozoans are unicellular heterotrophs which feed on a variety of organisms, including bacteria, algae, detritus, yeasts and in some cases, other protozoans (Horne and Goldman 1994). Some protozoans are obligate parasites on fish, phytoplankton or humans. The flagellates are generally considered to belong to the nanoplankton, while ciliates are classified as microplankton.

Rotifers are small, heterotrophic metazoans. They consume bacteria, algae, some ciliates and, in some cases, other rotifers (Bronmark and Hansson 1998). They are primarily parthenogenic, producing only females which are genetically identical to themselves, although some species may produce males in unfavorable environmental conditions (Wetzel 1983). The parthenogenic trait endows them with an ability for rapid reproduction allowing them to exploit transient periods of favorable conditions.

Two classes of crustaceans are important members of the zooplankton community: cladocerans and copepods. The diet of cladocerans consists of algae and bacteria, while that of copepods also includes zooplankton and detritus (Bronmark and

Hansson 1998). These crustaceans are classified as macrozooplankton, while the rotifers are classified as microzooplankton.

The crustacean *Mysis relicta* is a member of the zooplankton community mainly in cold, deep, oligotrophic lakes due to their sensitivity to low oxygen concentrations (Bronmark and Hansson 1998). *M. relicta* has a relatively varied diet that includes detritus, phytoplankton, rotifers, macrozooplankton (such as cladocerans and copepods) and benthic detritivores. In the presence of kokanee, they compete with this species for macrozooplankton. However, they have an advantage over kokanee in that they are capable of consuming a wider variety of prey organisms. *M. relicta* exhibits diurnal migrations, spending the daylight hours in deep, dark waters and rising close to the surface at night to feed (Spencer et al. 1991). Since kokanee feed in the metalimnion and upper hypolimnion region during the daylight hours, *M. relicta* only becomes a significant prey item for them when hydrologic conditions disrupt the mysid diurnal migrations.

While a few species of aquatic insects are planktonic or surface-dwellers, most aquatic insects are benthic (Bronmark and Hansson 1998). The diet composition of the different aquatic insects is highly variable, including detritus, algae, macrophyte tissue, invertebrates and even fish. This group of organisms is an important source of food for fish. Aquatic insects and large crustaceans such as *M. relicta* are members of the macrozooplankton class.

### *Fish*

Kokanee (*Oncorhynchus nerka*) and rainbow trout (*O. mykiss*) were the target species of the fertilization projects in both the Arrow Reservoir and Kootenay Lake. While rainbow trout and kokanee are both important sport fish, kokanee are also important as the major food source of the large piscivores, such as rainbow and bull trout.

The primary target species of the Arrow fertilization project is kokanee, a non-anadromous sockeye salmon which do not migrate to sea. Like anadromous sockeye, most kokanee are semelparous and spawn in their natal streams, but some may spawn in gravels along the shoreline of lakes when the conditions are ideal. When the eggs hatch in a spawning stream, the fry migrate downstream to a lake where they remain for the

remainder of their lives, returning to their natal stream to spawn and die. Kokanee generally live 3 to 5 years with most maturing at age 4 (Ford et al. 1995).

When kokanee fry first arrive in the lake they often spend a few months feeding within the shallow littoral zone before migrating to the pelagic zone, where they remain as adults.

All age classes of kokanee feed primarily on crustacean zooplankters. Fry, however, tend to eat smaller micro-zooplankton and benthic invertebrates obtained while they inhabit the littoral zone. Kokanee tend to exhibit diel crepuscular vertical migrations (Ford et al. 1995). They spend most of their time in the hypolimnion where the water is cooler and food is scarce. However, at dawn and dusk they migrate to the nutrient rich metalimnion and epilimnion to feed. While this vertical migration ceases after fall turnover, crepuscular feeding habits are maintained. Kokanee are preyed upon by a variety of piscivores, including rainbow trout. The young of these predators are often competitors of kokanee.

All rainbow trout spawn in small streams. Some populations migrate to rivers or lakes while others remain in their natal stream for their entire life cycle. Of those that migrate to lakes, if the lake is large with sufficient food (eg. kokanee), they can become relatively large piscivores as adults. In smaller lakes, the adult rainbow trout tend to be smaller and feed primarily on insects. The rainbow trout in Kootenay Lake and Arrow Reservoir are piscivores, feeding primarily on kokanee. Young rainbow trout tend to feed on large zooplankton, benthic invertebrates and terrestrial insects. (Ford et al. 1995)

The distribution of rainbow trout within a lake depends primarily on water temperature, dissolved oxygen concentration and the abundance of food. In general, they tend to be found in waters where the temperature is less than 18 °C and the dissolved oxygen concentration is greater than 3 mg/L (Ford et al. 1995).

Rainbow trout generally mature at 3 to 5 years of age, while Gerrard stocks, a unique strain of rainbow trout found in Kootenay Lake, often mature at 5 to 6 years. Ford et al. (1995) report that iteroparity is common in rainbow trout. In their literature review of BC sport fish they found that survival of rainbow trout after spawning is relatively low and repeat spawners have been reported to make up approximately 10 to 33 % of a population.

### *Aquatic community interactions*

Within an aquatic ecosystem all organisms belong to a food web in which they are linked together by interactions such as herbivory, predation, parasitism, competition and symbiosis. Only primary producers, bacteria, some fungi and a few protozoans and dinoflagellates are capable of directly utilizing the dissolved nutrients in a lake (Horne and Goldman 1994). These organisms therefore form the base of the food web by converting the aqueous nutrients and dissolved organic carbon (DOC) into a form of organic carbon that other organisms can utilize.

Primary producers utilize phosphorus and nitrogen primarily in the forms of phosphate and ammonia, respectively. These are assimilated along with CO<sub>2</sub>, various micronutrients and energy from the sun to create new organic cellular material. In addition to inorganic nutrients, such as nitrogen and phosphorus, bacteria utilize soluble organic matter (DOM) and detrital particulate organic matter (POM) as a food source (Wetzel 1983). Phytoplankton exudates, animal excretion products and microbial decomposition products each contribute to the soluble and particulate matter available to heterotrophs.

Even in an ecosystem with relatively few species the food web can be very complex. However, by grouping similar species into functional groups a much simpler representation of the ecosystem (or a part of it) can be created. For example, the organisms in the pelagic region of a lake can be represented by a food chain, in which phytoplankton feed zooplankton, which feed planktivorous fish, which feed piscivores. This may be represented as follows:

phytoplankton => zooplankton => planktivorous fish => piscivores

Classic food webs or food chains such as this one are, however, often incomplete as they do not include the microbial components of the ecosystem. More recently, limnologists have begun to better understand the importance of microbial food webs (MFW) in nutrient recycling and carbon flows in pelagic systems (Stockner and Porter 1988). It is important to include the microbial community in a food web, as they are responsible for recycling a significant quantity of nutrients, which would otherwise be lost. The main components of the microbial loop are bacteria, autotrophic picoplankton, heterotrophic flagellates and ciliates.



The base of the microbial food web consists of the hetero- and autotrophic picoplankton, which utilize DOC and  $\text{CO}_2$ , respectively, along with inorganic nutrients, such as nitrogen and phosphorus. These organisms are the primary food source for hetero- and mixotrophic nanoflagellates. The diet of ciliates consists of flagellates and picoplankton and these small microzooplankton are in turn consumed by larger micro- and macrozooplankton. All members of the food web release DOC, inorganic nutrients and  $\text{CO}_2$  as waste products or through inefficient feeding. These waste products may be re-utilized by the picoplankton and this cycling of nutrients within the microbial food web is referred to as the microbial loop.

An example of a pelagic food chain, including the microbial food web, is depicted in Figure 2.1.

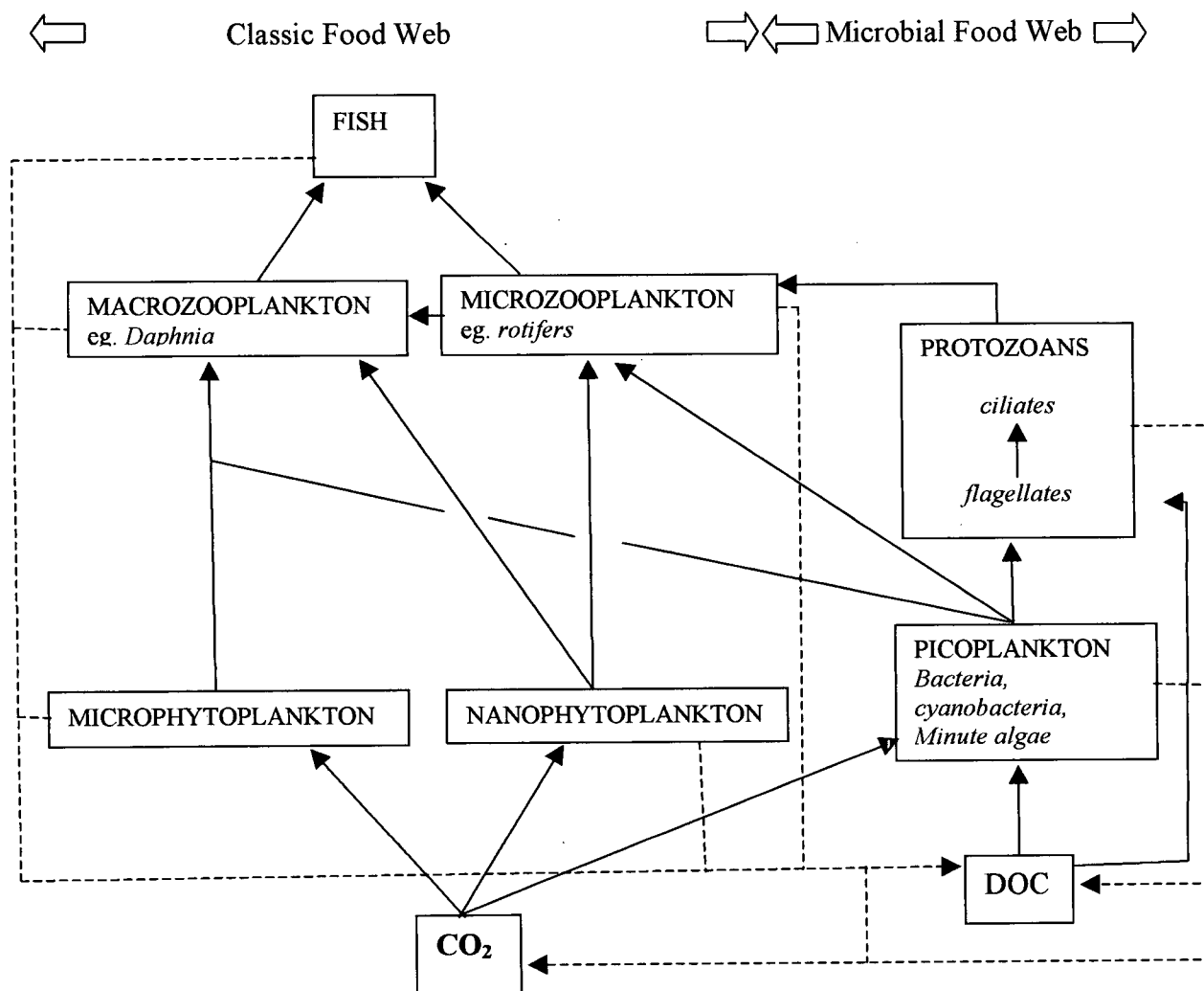


Figure 2.1. Schematic of the cycling of carbon within an aquatic ecosystem. Solid lines point from a food source to the consumer of that food source. The classic food chain is shown on the left and the microbial loop is depicted on the right. The carbon source for the classic food chain is CO<sub>2</sub>, while DOC is a major source of carbon for the microbial loop. Dashed lines represent the release of CO<sub>2</sub> and DOC by all members of the aquatic ecosystem, which contributes to the overall pool of organic and inorganic carbon.

The picophytoplankton contribution to primary productivity is greater in nutrient poor systems (Stockner and Antia 1986, Stockner and Shortreed 1989), such as the Arrow Reservoir. Consequently, the microbial loop plays a greater role in oligotrophic

than meso- or eutrophic lakes and reservoirs (Stockner and Antia 1986, Horne and Goldman 1994). In nutrient enriched systems, where the phytoplankton community is dominated by larger species, there are fewer steps in the transfer of energy from primary producers to fish. The dominant phytoplankton species of oligotrophic systems are not suitable prey for larger zooplankton. In these systems, energy is transferred from picoplankton to nanoplankton, microzooplankton and fish. Due to the substantial loss of energy with each step, the transfer of energy from primary producers to fish is less efficient in picoplankton based food webs. In oligotrophic systems, the recycling and retention of DOC, nitrogen and phosphorus within the epilimnion by the members of the microbial loop, which, in general, do not sink, is vital for maintaining the productivity of the ecosystem.

### **2.1.3 Reservoirs and their effects on aquatic ecosystems**

Reservoirs are created to store water for a variety of uses, including hydropower generation, flood control, industrial and municipal water use, irrigation, navigation, fishing and recreation. Often, many of these uses coincide within the same reservoir. The nature and operation of a reservoir will depend on the primary use(s) of the stored water. A reservoir may be created using an existing natural lake or a completely artificial lake may be created through the impoundment of a river basin. They usually are dendritic or longitudinal in shape and have a high shoreline development ratio (Benson 1982).

Reservoirs differ from natural lakes in many respects. Reservoirs generally have a greater drainage to surface area ratio and thus a larger potential sediment and nutrient load (Benson 1982). The water level fluctuations in reservoirs are often relatively high and the seasonal patterns are generally different from those in natural systems. This has severe consequences for the biotic communities within the reservoir as well as those downstream of the reservoir. In addition, the nature of the morphology and hydrology of reservoirs favours gradients in the biotic and abiotic characteristics of the reservoir, which are generally more well defined than in natural lakes (Kennedy et al. 1982, Van Den Avyle, M.J. et al. 1982).

The creation of a reservoir will have some impact on the local aquatic physical, chemical and biological characteristics. The local terrestrial biotic and abiotic environment may also be affected. The nature and severity of these impacts are highly variable and are dependent on many parameters, including the structure, size and operation of the dam, as well as local climate, geology and hydrology. (Sundborg, 1982)

In addition to local impacts, there may be significant impacts to ecosystems which are relatively far from the reservoir. The effects of the construction and operation of the WAC Bennett Dam on the upper Peace River in BC on the Peace-Athabasca Delta in Alberta is a good example of this (Healy and Wallace 1987). The Peace River, which originates in BC is a major tributary of Lake Athabasca in Alberta. Prior to the construction of the WAC Bennett Dam the annual flooding of the Peace-Athabasca Delta sustained an early successional, highly productive vegetative cover within the delta. The flooding also resulted in a periodic connection to perched basins providing a supply of water to these basins. The annual flooding ended following the closure of the dam due to the decreased water levels in the Peace River. In the absence of flooding, vegetational succession proceeded and meadow and willow ecosystems formed. In addition, the perched basins began to dry out and wildlife, such as muskrats, ducks and walleye, was adversely affected.

Within a reservoir, the changes that take place following the closure of the dam usually follow the same general pattern, often referred to as "boom and decline". Following reservoir filling, nutrients and detritus are released by the flooded terrestrial vegetation and soils. These additional nutrients promote increased plankton and fish production. Over time, this additional nutrient source is depleted and the tributaries once again become the primary source of nutrients to the reservoir. As the nutrient and detrital input of the flooded region declines, the productivity of plankton and fish decreases and levels off, often at a level similar to that prior to impoundment. Benson (1992) reports that the production levels off in five to ten years. However, Stewart (1982) reports that stabilization is generally achieved sooner in reservoirs created from existing lakes than in completely man-made lakes, where it may take 30 or more years to reach equilibrium.

The flooding of terrestrial soils also results in increased cellular concentrations of methylmercury (MeHg) in zooplankton and fish (Patterson et al. 1998). Mercury (Hg)

accumulates in the humic horizon of forest soils due to the deposition of atmospheric Hg (Montgomery et al. 1995). When these soils are flooded, bacterial methylation of inorganic Hg is stimulated by the decomposition of organic matter in the recently flooded region. The methylated form of mercury has the potential for bioaccumulation and biomagnification in food webs via macrobenthic food chains.

As previously mentioned, the relatively large water level fluctuations in a reservoir may have several deleterious effects within the reservoir. Erosion of the region between the maximum storage level and the minimum drawdown level in a reservoir is often greater following impoundment. This generally results in an increase in turbidity and sediment deposition to the benthic zone (Sundborg 1982, Healy and Wallace 1987). Increased turbidity may affect primary productivity, while increased sediment deposition may alter benthic habitat (Liao et al. 1988). Large, untimely water fluctuations may also desiccate the spawning locations of some fish species, killing any developing eggs which are present (Modde et al. 1997). In addition, changes in the timing and magnitude of water level fluctuations may decrease or eliminate the littoral vegetation (Sundborg 1982).

The altered hydrology, littoral habitat and nutrient status within the newly formed reservoir may alter the species composition and interactions. Benson (1982) reports that while riverine and lacustrine fish thrive initially following impoundment, a reduction in the number of fish species and biomass may occur. Changes in the species composition of fish (Benson 1982, Roy 1982, Stewart 1982, Liao et al. 1988, Crivelli et al. 1995), zooplankton (Pinel-Alloul et al. 1982, Roy 1982, Ioriya 1998) and phytoplankton (Zakova et al. 1993, Ioriya 1998) are often observed following impoundment.

The local groundwater regime may be affected to some extent by the construction and operation of a dam. Since the groundwater table is dependant on the water level in the reservoir, fluctuations in the water level may affect the local vegetation (Sundborg 1982).

The impacts of a reservoir on the downstream environment are numerous. The sediment load is generally reduced downstream of a reservoir, altering the physical characteristics of the river. Changes to the shape of the river bed, such as channel incision and disappearance of mid-channel bars and islands, as well as changes in the

substrate may drastically alter aquatic habitat (Ligon et al. 1995). Changes at the expense of one organism may be beneficial to others.

Reservoirs generally decrease downstream flow variation and minimize or eliminate downstream flooding. This has a significant impact on fish which utilize the inundated flood plains during a portion of their life cycle (Ligon et al. 1995, Power et al. 1996). Artificially regulated flows may also alter successional patterns of primary producers and consumers, which may affect the food web dynamics (Power et al. 1996). It has been suggested that regulated flows which differ significantly from the natural flows reduce biodiversity and alter predator-prey dynamics (Power et al. 1996).

Upon filling a reservoir, the decomposition of organic matter may deplete the dissolved oxygen near the bottom resulting in anaerobic conditions (Ioriya et al. 1998). In reservoirs which stratify thermally, the lack of reaeration in the hypolimnion may result in a significant decline in the water quality of this layer due to low oxygen concentrations, elevated concentrations of the reduced forms of iron and manganese and the production of hydrogen sulfide (Symons et al. 1964). Release of water such as this or highly turbid water from the reservoir may have significant impacts on the downstream water quality. High concentrations of iron and manganese must be removed from drinking water reservoirs (Zaw and Chiswell 1999).

The water temperature regime downstream of a reservoir may be affected by the reservoir and this in turn may affect the aquatic ecosystem. Webb and Walling (1993) report an increase in mean water temperature, a decrease in summer maximum water temperature and diel fluctuations, a delay in the seasonal regimes and the elimination of ice formation. Salmon spawning, rearing and migration may be affected by the release of warm epilimnetic water from a stratified reservoir (Ruggles and Murray 1983). Camargo and Voelz (1998) report changes in macroinvertebrate communities exposed to hypolimnetic release from reservoirs on the Colorado (USA) and Duraton (Spain) Rivers, which are used for water storage and hydroelectric production, respectively. The factors responsible for these changes were cooler summer and warmer winter water temperatures in the Colorado River and flow fluctuations and low dissolved oxygen concentrations in the Duraton River.

Air entrapment and supersaturation may occur below some dams. When fish, which are at equilibrium with the supersaturated water, migrate to areas having lower atmospheric pressure, gas bubbles form inside their bodies resulting in gas embolism and death (Ruggles and Murray 1983). Of the atmospheric gases, nitrogen plays the most significant role in gas embolism as it has the highest partial pressure.

Many measures have been proposed to minimize the impacts of the construction and operation of a reservoir. The migration of fish past dams is often aided by fishways or mechanical transportation methods, such as fish lifts (Sundborg 1982, Stewart 1982). Water level fluctuations may be managed with consideration for the requirements of the resident or anadromous fish species (Benson 1982). However, operations, such as the maintenance of stable full pool water levels throughout the growing season, which benefit resident fish may be harmful to anadromous fish. Likewise, operations, such as flow augmentation, which benefit anadromous fish may have negative impacts on resident fish. (Geist et al. 1996)

Shoreline protection measures are often applied in reservoirs to prevent erosion (Sundborg 1982) and vegetation is occasionally planted on exposed shorelines when the water level is low to provide littoral habitat (Benson 1982). Larson (1982) reports that where water quality varies with depth, some of the effects on water quality within the reservoir and downstream may be minimized or eliminated in reservoirs which are capable of selectively withdrawing water at one of several well-spaced depths. Mechanical destratification measures may be considered for some reservoirs which suffer from problems related to low dissolved oxygen concentrations in the hypolimnion (Benson 1982, Zaw and Chiswell 1999).

In the past, some reservoirs have been filled without any deforestation of flooded regions. This flooded vegetation contributes to the increased biochemical oxygen demand of the flooded regions and may become a nuisance or a hazard for recreational users. However, complete clearing may increase the potential for erosion in the basin and flooded vegetation provides habitat for aquatic biota. Kiell (1982) documented a selective clearing strategy for the Upper Salmon Reservoir in Newfoundland to minimize erosion, reductions in dissolved oxygen and impacts to recreational users and to provide

additional fish habitat. Selective clearing was also used in the lakes of the Tennessee-Tombigbee Waterway to provide fish habitat (Sims 1982).

The design and operation of a reservoir should reflect the various users within and surrounding the reservoir, as well as the local ecosystem. Since the water requirements of the various users, including the local biota, are often conflicting, a water management plan should be designed to minimize negative impacts and benefit the users to the greatest extent possible.

## **2.2 Relevant History of the Arrow Reservoir**

The Columbia River Basin, which spans portions of western Canada and the United States, is shown in Figure 1.1. In 1964, the governments of these two countries approved the Columbia River treaty, which specified that three storage reservoirs were to be constructed in the Canadian section of the Upper Columbia River Basin. The dams were to be built at Mica Creek and in the Duncan and Arrow Lakes with the mandate of providing increased power generation and flood control in the Columbia River Basin (Forrest 1978).

The Keenleyside Dam was completed in 1968 on the Lower Arrow Lake near Castlegar and, in 1973, the Mica Dam was completed on the Upper Columbia River. In addition, in 1984, the Revelstoke Dam (which was not connected with the Columbia River treaty) was finished near Revelstoke at the inlet to the Upper Arrow Lake (Pieters et al. 1998).

The effects of the construction and operation of these dams on the Arrow lakes fish populations have been profound. Immediate, direct effects include the inhibition of fish migration and flooding of spawning and rearing habitat. Following impoundment, an estimated 30% of the Arrow lakes basin spawning and rearing habitat was lost due to flooding (Lyndsay and Seaton 1978) and migrations south of the lakes were impeded. The construction of the Revelstoke dam created a physical barrier to north-bound migrations of Arrow lakes fish and resulted in the flooding of approximately 150 km of the mainstem Columbia River and 200 km of its tributaries (Pieters et al. 1998). Relatively few Arrow Lakes fish are believed to have migrated past the location where the Mica Dam was built. Therefore, the direct impacts of the Mica Dam on the Arrow



lake's fish due to flooding and the erection of a physical barrier are considered minimal. However, kokanee did spawn above the location of the Mica Dam contributing to the general nutrient supply of the system.

Prior to construction of the Revelstoke dam, the impending impacts of this project on fish populations and habitat were anticipated by those involved in the construction of this dam. Therefore, funding to replace losses of fish and fish habitat was secured with BC Hydro and the BC Ministry of the Environment signing a compensation agreement. A portion of this funding was used for the construction of the Hill Creek Spawning Channel and the Hill Creek Hatchery during the periods of 1979-1980 and 1982-1983, respectively.

Despite these efforts to enhance the Arrow lakes fish populations, data from trawl and hydroacoustic surveys, kokanee spawner enumerations and sport harvest estimates in the decade following Revelstoke dam construction indicated that fish populations were declining (Pieters et al. 1998). The primary cause of the rapid decline of the fish populations is believed to be the oligotrophication of the Arrow Reservoir. The main factors contributing to this are the retention of nutrients by upstream impoundments, the lack of littoral vegetation and the altered hydrograph.

An additional problem for some of the Arrow Reservoir fish populations, particularly kokanee, is the presence of *Mysis relicta*. This freshwater shrimp was introduced into the reservoir in 1968 and 1974 with the intention of providing and additional food source for kokanee (Lasenby et al. 1986). However, the mysids proved to be efficient at avoiding predation by larger kokanee, while competing with both juvenile and adult kokanee for food resources.

### **2.3 Previous Lake Fertilization Projects**

In general, the ultimate goal of lake fertilization is to increase resident fish growth and survival and restore populations to historic levels. It has been well documented (Stockner and MacIsaac 1996) that the addition of nutrients to BC sockeye rearing lakes results in larger increases in primary productivity ( $>2X$ ). In theory, if the N:P ratio and the load is optimum then primarily the smaller species of phytoplankton, which are a suitable food source for zooplankton, will be stimulated. This in turn should enhance the

productivity of subsequent desired trophic levels as has been demonstrated (Stockner and MacIsaac 1996).

Lake fertilization has been performed in Europe (Jansson 1978, Lundgren 1978, Bjork-Ramberg and Anell 1985, Australia (Weatherley and Nicholls 1955) and North America (Gross et al. 1996, Stockner and MacIsaac 1996, Ashley et al. 1997, Johnston et al. 1999). Since lake fertilization has been studied extensively in the province of British Columbia, this section will focus on these studies.

The majority of lakes in the coastal region of BC are oligotrophic or ultra-oligotrophic due to the flushing of available nutrients out of the system by frequent, heavy rainfalls. Populations of anadromous pacific salmon spawn in the tributaries to these lakes and the decaying carcasses of the spawned salmon provide a source of nutrients to the lakes. However, heavy fishing of the salmonids which spawn in these coastal regions has restricted this nutrient supply. This phenomenon of carcasses of spawned anadromous salmonids providing nutrients to their natal streams is referred to as the anadromous nutrient pump.

Following a promising study in the early 1970s on the effects of fertilization on sockeye salmon (*Onchorhynchus nerka*) productivity in Great Central Lake, BC (LeBrasseur et al. 1978) a Lake Enrichment Program began in the province in 1977 (Hyatt and Stockner 1985; Stockner and Shortreed 1985; Stockner and McIsaac 1996). The goal of this program was to assess the effectiveness of lake fertilization as a method of enhancing sockeye salmon production to compensate for the partial loss of the anadromous nutrient pump. A total of 20 oligotrophic coastal BC lakes were fertilized with varying frequencies. The N:P ratio and the load varied from lake to lake and were designed for the local conditions (physical, hydrological and biological) of each lake.

Increases in biomass were observed in fertilized lakes in varying amounts at all trophic levels. Significant increases were observed in the following: bacterioplankton abundance, autotrophic picoplankton abundance, chlorophyll concentration, phytoplankton volume, primary production, zooplankton biomass and fish smolt weight. Responses to fertilization varied from lake to lake depending on local variables. Increases in productivity were lower in treated glacial lakes due to light limitation resulting from the high turbidity in these lakes, which is due to a high load of glacial

flour. In some lakes, competition by threespine sticklebacks (*Gasterosteus aculeatus*) has reduced the effects of fertilization on fry weight (Stockner and Hyatt 1984). However, in general, yearling smolts were significantly larger in treated lakes. In addition, where the data are available, it appears that returns are significantly greater in fertilized lakes than in untreated lakes (Stockner and Hyatt 1984).

A cost/benefit analysis revealed that the value of the additional salmon returns more than exceeds the cost of fertilization. In addition, of all current salmonid enhancement techniques, lake fertilization has the lowest cost:benefit ratio (Stockner and McIsaac 1996).

In addition to studying the effects of lake fertilization, the recovery of lakes following the termination of fertilization was assessed. The return of additional salmon and the decay of their carcasses following spawning should theoretically maintain an enhanced salmon population. However, an active fishery harvests an estimated 60 to 70 percent of returning salmonids, removing this nutrient source from the system. Following termination of lake fertilization the nutrients, primary productivity and zooplankton biomass reached conditions similar to those prior to treatment (Stockner and McIsaac 1996).

In addition to nutrient availability (bottom up control), the biomass at a given trophic level is a function of birth rates and mortality. Predation (top down control) may play a major role in controlling the biomass of a group of organisms. Since parameters such as competition, predation and the number of offspring produced may be a function of size in some species, the response to lake fertilization may be more complex than simple increases in size or abundance. Johnston et al. (1999) performed a long term lake fertilization experiment to study the relative importance of bottom up and top down control mechanisms on biomass at various trophic levels. A before-after control-impact experimental design was used in which West Twin Lake served as a control and East Twin Lake was fertilized for a 5 year period from 1990 to 1994. These oligotrophic lakes, which are located in the Coast Mountains of BC, were studied for 8 years (1982-1989) prior to fertilization to provide background data.

Phytoplankton, zooplankton and zooplanktivorous rainbow trout constitute the three main trophic levels in the pelagic region of the lakes. Increases in biomass were

observed at all trophic levels in response to fertilization. While the biomasses of phytoplankton and zooplankton were directly proportional to average total phosphorus (TP) concentrations in the lake, the biomass of rainbow trout varied as  $TP^{0.5}$ . This supports the hypothesis that bottom-up effects are reduced as they are transmitted along a food chain. The results of this study indicate that the role of top down effects in controlling biomass was minimal in the oligotrophic Twin Lakes.

The study of greatest relevance to the fertilization of the Arrow Reservoir is the Kootenay Lake Fertilization project. Kootenay Lake is located in the southern interior of BC to the east of the Arrow Reservoir. This lake is very similar to the Arrow Reservoir in many respects. Located in the Purcell mountain trench, Kootenay Lake is a long, narrow, deep fjord-like lake. It is naturally oligotrophic and is influenced by dams at its major inflows (the Duncan Dam was built on the Duncan River in 1967 and the Libby Dam was built on the Kootenai River in 1972) and its outflow (the Corra Lynn Dam was built in 1931; 4 other dams also have been built in between Nelson and Castlegar).

A variety of species of fish inhabit Kootenay Lake, including several sportfish. The sportfishery is focussed primarily on kokanee, rainbow trout and bull trout. A distinct spawning stock exists in each of the three arms of the lake (North Arm, West Arm and South Arm). There are also two distinct strains of rainbow trout: the larger Gerrard strain and the smaller "resident" rainbow. In 1990, the annual value of the Kootenay Lake fishery was estimated to be \$1,136,800 (Thompson 1999).

Kootenay Lake has suffered numerous human-inflicted perturbations/disturbances in the past 5 decades. In 1949 and 1950, *M. relicta* was transplanted into this lake from Waterton lake in Alberta (Sparrow et al. 1964). This became the site of the first successful transplant of this species when, in the late 50's and early 60's, it became apparent that the population had established itself. The purpose of the introduction of this species of shrimp was to provide a food source for young rainbow trout at the life stage where they switch from zooplankton to kokanee. Evidence suggests that the mysid population was growing up until the early 1980s (Lasenby et al. 1986).

In 1953 a fertilizer plant began operation near Kimberly BC. With no environmental controls in place, nutrients were released directly into St Mary River, a tributary of Kootenay River, upstream of Kootenay Lake. However, pollution controls

were subsequently installed with a settling pond in 1969 and with effluent recycling in 1975 (Thompson 1999).

As previously stated, Duncan River and Kootenay River were impounded in 1967 and 1972, respectively. Both of these dams are used for water storage and flood regulation, while the Libby Dam is also used for power generation. The operation of these dams has altered the flow regime in Kootenay Lake. The spring and summer flows from both of these rivers are significantly lower and the winter flows are much higher than historical flows prior to impoundment. In addition, the suspended solids and nutrient loads to the lake decreased significantly following impoundment of the main inflows (Thompson 1999).

The combination of mysid introduction, pollution and the impoundment of the main inflows of Kootenay Lake has had a major impact on the aquatic ecosystem. In the 1960's the mysid population had established itself and the phosphorus load from the fertilizer plant increased following a doubling and then tripling of production in 1962 and 1964, respectively. Increased growth rates were observed or inferred in rainbow trout in the 1950's and 1960's (Lasenby et al. 1986). Since mysids were not a major component in the diet of these fish, this is likely due to the eutrophication of Kootenay Lake during this period. In the 1960's kokanee in the West Arm of the lake and at the mouths of some tributaries were significantly larger than in the past. In addition to the increased productivity during this time, the presence of mysids is a major factor in the increased size of these kokanee. The West Arm is relatively shallow and experiences relatively high flows and these hydrologic conditions cause mysids to be more vulnerable to predation by kokanee (Martin and Northcote 1991). There was no evidence to suggest that kokanee in the main lake were larger during this period. In fact, it is likely that the eutrophication of the lake during this period compensated for the increasing mysid competition, which would otherwise have been detrimental to this kokanee population. It should be noted here that a mistaken belief that the presence of mysids enhanced rainbow trout and kokanee populations in Kootenay Lake provided the rationale for the introduction of this species to the Arrow Reservoir.

Following the impoundment of the main inflows and the implementation of pollution controls at the fertilizer plant, the nutrient load to Kootenay Lake plummeted.

In 1977, the orthophosphate load to the lake was one third of the historical load (Daley et al. 1981). The effects of this oligotrophication on the ecosystem became apparent in the mid 1980's when the annual abundance of kokanee spawners began decreasing steadily. This alarming trend was apparent in all stocks, with the exception of those in the west arm.

The decline of the Kootenay Lake kokanee was attributed to a combination of the oligotrophication of the lake and competition from the mysids. There is currently no known method for removing mysids from a lake in which they have become established. In the late 1980's lake fertilization was proposed as perhaps the only method for preventing the collapse of the kokanee followed inevitably by the collapse of the rainbow trout.

In 1991, a lake fertilization response model was devised for Kootenay Lake (Walters et al. 1991). Contradictory results were obtained when this computer model was run with different variations of the parameter estimates. In one scenario the model predicted that the mysids would benefit from the fertilizer at the expense of the kokanee. In this scenario, the crash of the kokanee population was accelerated by fertilization. The model also predicted that the kokanee stocks would crash even in the absence of fertilization. With no other options, the decision was made to begin a fertilization program in Kootenay Lake.

An intensive 5 year fertilization program was implemented in Kootenay Lake in 1992 (Ashley et al. 1997). The main objective of this program was to restore the kokanee population by reversing the oligotrophication of the lake. From 1992 to 1996 fertilizer was added to the lake and extensive monitoring of the ecosystem was carried out. Since 1996, the lake fertilization and monitoring have continued at a lower level. As with the Arrow Lakes project, a period of 5 years was chosen for the initial study in order to encompass an entire kokanee life cycle (4 years) plus one additional year.

The results of this study have been promising with respect to most biological and chemical features of the ecosystem which were studied. It was feared that the additional nutrients may benefit the mysids at the expense of the kokanee. However, the mysid population decreased during the first four years of fertilization from 405 individuals/m<sup>2</sup> to 126 individuals/m<sup>2</sup> and then increased slightly to 195 individuals/m<sup>2</sup> in 1997. For the

sake of comparison, the highest annual average mysid densities were in 1987 when 745 individuals/m<sup>2</sup> were observed. Mysids appear to be adversely affected by high water flow through the lake and during seasons of high flows their abundance tends to be low. During the first year of fertilization the flow was relatively low, however, the flows through the lake increased throughout the fertilization study period.

As previously stated, the main goal of this project was to restore kokanee biomass and productivity to historical levels in Kootenay Lake. The results from the first five years of fertilization indicate that the study has been relatively successful with respect to the kokanee populations (Thompson 1999). Kokanee spawner abundance increased significantly in the North Arm during the study. While this parameter also increased in the West Arm, the trend began several years prior to the initiation of fertilization. In the South Arm, kokanee spawner abundance did not appear to be affected by the fertilization, which was carried out in the North Arm. Historically, this population has been significantly lower than the North and West Arm populations. The total abundance of kokanee within the lake has also increased significantly during the study.

The rainbow trout populations in Kootenay Lake also appear to have benefited from the fertilization project. Spawner abundance increased during the project and this trend continued in 1998 and 1999 (Thompson 1999).

Phytoplankton and macrozooplankton biomass increased during the study. Historical microzooplankton abundance data is not available and the data from this study indicates that this parameter did not change significantly during the study.

## **2.4 Arrow Reservoir Fertilization**

The addition of fertilizer to the Arrow Reservoir began in 1999 at the start of the freshet in April and continued for 20 weeks until the end of August. The expected phosphorus concentration in the epilimnion and the N:P ratio of the fertilizer during the first week of fertilizer addition (April 19 to 23) were 1.1 ug/L and 0.67 (wt:wt), respectively. Over the course of the fertilization period these variables both gradually increased such that during the final week of fertilization (August 30 to September 3) the epilimnetic phosphorus concentration was 6.5 ug/L and the N:P ratio was 7.5. In addition, the frequency of fertilizer addition increased from 2 times per week during the

first week to a peak of 15 times per week during the week of August 2 to August 9. (Ken Ashley, personal communication)

Fertilization was achieved using the Galena Bay Ferry, which crosses the reservoir perpendicular to the flow at the north end of the upper basin. Large 3800 L tanks were trucked onto the ferry and the fertilizer was pumped into the lake via a diffuser unit which was bolted to the rubbing streak of the ferry. This allowed the fertilizer, which is significantly more dense than water, to be well dispersed into the surface waters of the reservoir. The diffuser unit was a 7.62 cm (3 inch) perforated stainless steel pipe with 15.24 cm (0.25 inch) holes every 0.635 cm (6 inches) of the entire length.

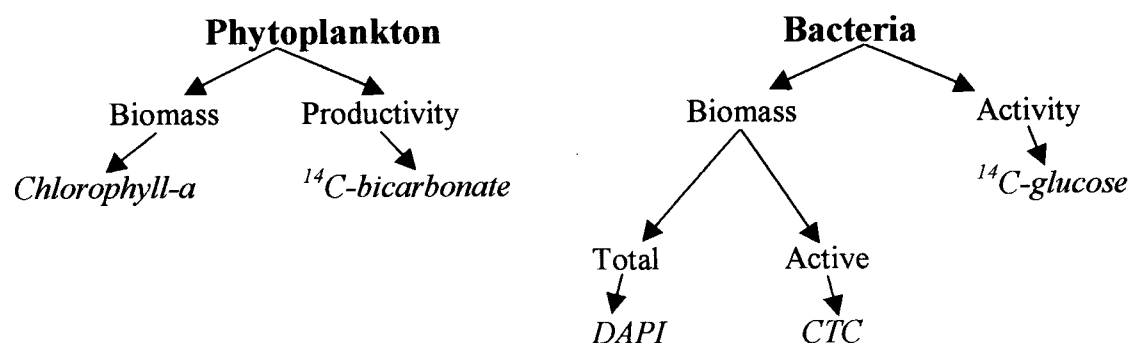


### 3. METHODOLOGY

#### 3.1 Vertical Transects

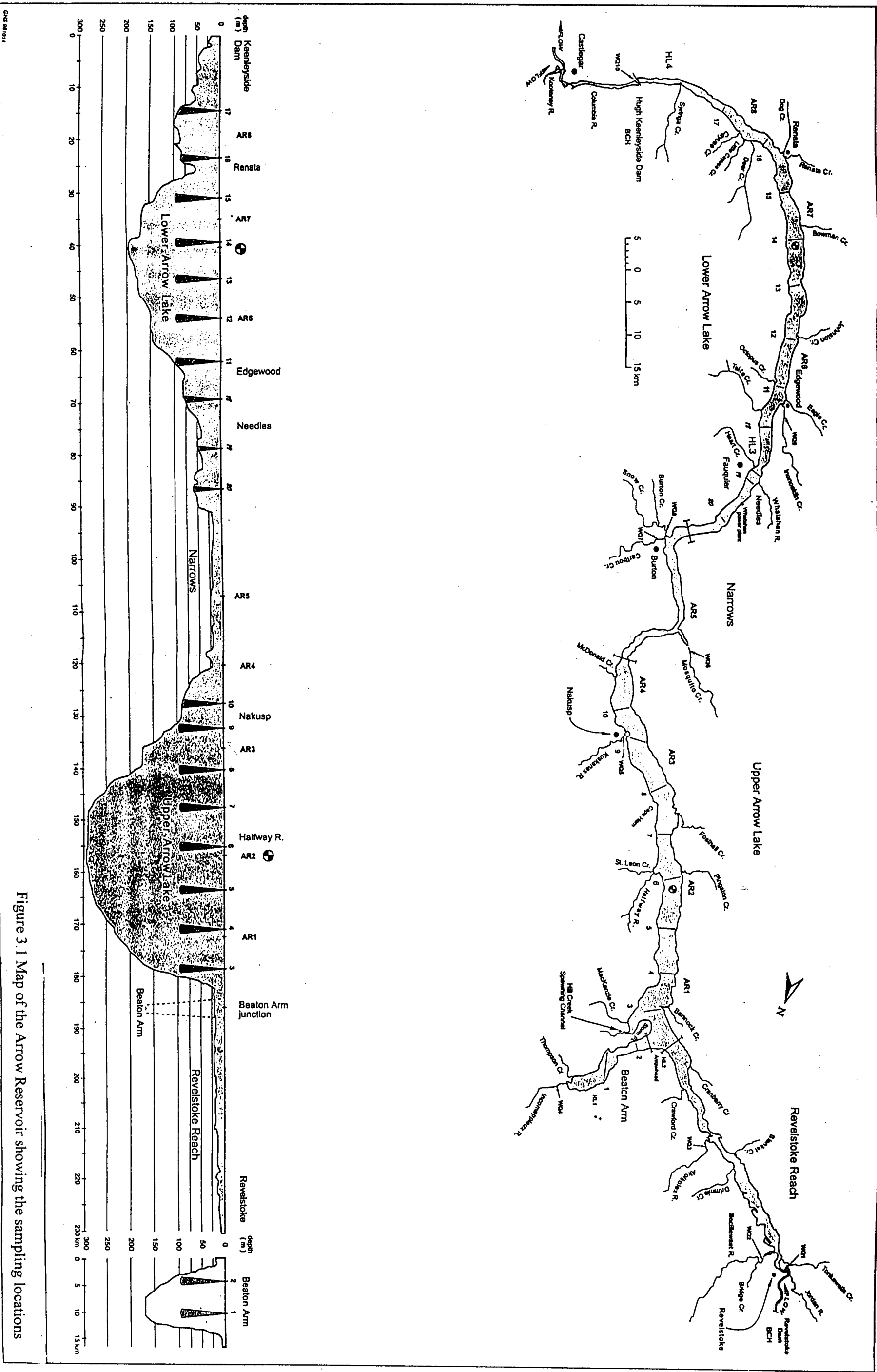
Vertical transects were performed in both the upper and lower basins of the Arrow Reservoir on a monthly basis from May to September in 1998 and from April to September in 1999. These transects, which provide a profile of the phytoplankton and bacterial communities from the surface to a depth of 15 m, were performed at a single central location in each lake (shown in Figure 3.1). Samples were collected from six depths at each location and several experiments were performed to estimate the algal and bacterial biomass and productivities.  $^{14}\text{C}$ -bicarbonate incorporation experiments and chlorophyll-a analyses were used to estimate primary productivity and algal biomass, respectively. Estimates of bacterial activity were provided by and  $^{14}\text{C}$ -glucose incorporation studies. DAPI and CTC counts were used to measure bacterial abundance (biomass) and the percentage of active bacteria, respectively. Figure 3.2 depicts a summary of the experiments performed.

Figure 3.2 - Summary of vertical transect experiments



In June of 1999, the lower basin was not sampled due to problems with the boat trailer. In addition, lower basin samples were collected close to the boat launch in August due to motor problems. No incubations for productivity were performed on the lower lake on this trip and only chlorophyll-a, DAPI and CTC samples were collected.

A van Dorn sampler was used to collect samples from depths of 1, 2, 5, 10 and 15 m at each sample location. In addition, samples were collected directly from the surface.



These depths were chosen to provide an adequate profile from the surface to a depth which corresponds to approximately 1% light penetration. Surface light penetration was measured with LiCor quantum meter (model 185A) at 1 m intervals from the surface down to at least 1% light. The secchi depth was also measured on each sampling occasion.

Samples which were not processed immediately were transported to the University of British Columbia Environmental Engineering laboratory on ice.

### **3.2 Horizontal Transects**

Horizontal transects were performed in the upper basin in June, August and September of 1999. These transects were performed to provide a horizontal profile of the primary productivity in the Upper Arrow basin, from the Galena Bay ferry to Nakusp, at a depth of 2 m. This depth was chosen as it is approximately the depth of maximum primary productivity. Samples were collected from a depth of 2 m at six locations in the upper basin, shown in Figure 3.1. Station 1 is located at the north end of the basin, closest to the ferry where fertilizer is added, while station 6 is near the south end of the lake. Since the vertical transect was approximately mid-way between stations 3 and 4, the 2 m samples from the vertical transect were included in the horizontal transect series as station 3.5.

Samples were collected with a van Dorn sampler for chlorophyll-a analysis and  $^{14}\text{C}$ -bicarbonate incorporation. Estimates of bacterial concentrations and activities were not performed due to time and resource constraints.

### **3.3 Containment Experiments**

Containment experiments were carried out on the following occasions in 1999: mid-April, mid-May and mid-July.

Surface water was collected from the docks at the Nakusp marina and transferred into four 20 L plastic carboys. One of these carboys served as a control, while solutions of nitrogen (N) and phosphorus (P) in varying ratios were added to the other three. One carboy received a solution with an N:P ratio identical to that which was being added to the lake at the time the sample was collected. Of the other two carboys receiving

nutrients, one received a higher N:P ratio and the other received a lower N:P ratio. The final phosphorus concentration in each of the three carboys receiving nutrients was approximately the same as the estimated concentration in the epilimnion of the lake due to fertilizer addition. The N:P ratio and final phosphorus concentrations in each bag are presented in Table 3.1. In July, the lake water was filtered through a plankton net, to remove zooplankton prior to filling the carboys.

Table 3.1 – Containment experiment phosphorus concentrations and N:P ratios

| Sample period | N:P ratio |           |          | Expected Concentration of P (ug/L) |
|---------------|-----------|-----------|----------|------------------------------------|
|               | Carboy 1  | Carboy 2* | Carboy 3 |                                    |
| April         | 0.45      | 0.67      | 1.50     | 1.1                                |
| May           | 1.50      | 3.00      | 4.50     | 2.2                                |
| July          | 4.50      | 6.00      | 7.50     | 5.0                                |

\* Ratio added to the lake during the respective months.

Each of the four carboys was tied to a set of floats at the Nakusp marina, such that they were just submerged and were not shaded by the floats. The carboys were left to incubate for a period of approximately 3 days. Following the incubation, water from each carboy was used to fill three light and one dark BOD bottle for a  $^{14}\text{C}$ -bicarbonate incubation. In addition, water was transferred from each carboy into separate 500 ml plastic bottles for chlorophyll-a analysis.

In April, the samples were transported to the UBC Environmental Engineering laboratory on ice following the termination of the incubation. The  $^{14}\text{C}$ -bicarbonate incubations were performed in a temperature controlled room at 24 °C and saturating light. During May and July the  $^{14}\text{C}$ -bicarbonate incubations were performed *in situ* at the surface of the lake. The samples were preserved and transported to the UBC Environmental Engineering laboratory at the end of the incubation.

Each of the  $^{14}\text{C}$ -bicarbonate incubations was allowed to proceed for a period of four hours and then terminated with 1 ml of 37% formaldehyde. Samples were processed as described in section 3.5.1.

### 3.4 Stream Bioassays

The bioavailability of nutrients in several Arrow Reservoir tributaries was assessed using nutrient addition/omission tests. This nutrient bioavailability assay was designed based on the Environment Canada Environmental Protection Series *Selenastrum capricornutum* toxicity test (Environment Canada 1993). *Selenastrum capricornutum* was chosen as the test organism due to the fact that it is a standard organism used in toxicity testing with well-defined growth parameters. It is a small (40 to 60  $\mu\text{m}^3$ ) single-cell algae which does not form aggregates.

The goal of these tests was to determine whether nitrogen (N), phosphorus (P) or one or more micronutrients was limiting in the sample. This was done by measuring the growth of *S. capricornutum* in samples spiked with N, P or micronutrients (Nu) or a combination of the three. The hypothesis is that samples spiked with the limiting nutrient(s) should experience increased growth rates. The final concentrations of each of the nutrients in the algal incubation tubes, including the micronutrients are given in Table 3.2.

Table 3.2: Concentrations of the nutrients in the incubation tubes

| Nutrient | Source  | Concentration (mg/L) |
|----------|---|----------------------|
| N        | $\text{NaNO}_3$                                     | 2.52                 |
| Mg       | $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$           | 1.58                 |
| Ca       | $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$           | 0.72                 |
| S        | $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$           | 1.15                 |
| P        | $\text{K}_2\text{HPO}_4$                            | 0.11                 |
| K        | $\text{K}_2\text{HPO}_4$                            | 0.280                |
| Na       | $\text{NaHCO}_3$                                    | 6.58                 |
| C        | $\text{NaHCO}_3$                                    | 1.28                 |
| B        | $\text{H}_3\text{BO}_3$                             | 19.39*               |
| Mn       | $\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$           | 68.97*               |
| Zn       | $\text{ZnCl}_2$                                     | 0.94*                |
| Co       | $\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$           | 0.21*                |
| Cu       | $\text{CuCl}_2 \cdot 2\text{H}_2\text{O}$           | 0.003*               |
| Mo       | $\text{Na}_2\text{MoO}_4 \cdot 2\text{H}_2\text{O}$ | 1.7*                 |
| Fe       | $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$           | 19.8*                |

\* concentration is in  $\mu\text{g/L}$

NOTE:  $\text{Na}_2\text{EDTA} \cdot 2\text{H}_2\text{O}$  was present in each tube at 179.3  $\mu\text{g/L}$ .

Approximately 1 L of sample was collected from each tributary during the freshet in 1998 and then again during low flow periods in August of 1998. These samples were frozen and shipped to the UBC Environmental Engineering laboratory where they remained frozen until further processing.

Samples were thawed overnight prior to beginning the bioassay. Samples were assayed one at a time. Approximately one half of the sample was filtered and the bioassay was replicated on both filtered and unfiltered sample. Two ml of each of the following solutions was added in duplicate or triplicate to separate test tubes containing 20 ml of sample: N, P, Nu, N + P, N + Nu, P + Nu, N + P + Nu and distilled deionized water. One ml of *S. capricornutum* was then added to each test tube such that the final concentration was approximately 10,000 cells/ml. Samples spiked with N and P and micronutrients or distilled deionized water, but without *S. capricornutum*, served as controls.

These samples were incubated at 24 °C and saturating light for a period of 72 hours. The particles in each sample were counted with a coulter counter (TA II) and fluorometer (Turner 10-AU) readings were taken prior to the start of the incubation (time = 0 hours) and at 24, 48 and 72 hours into the incubation. Net planktonic growth was estimated based on the difference between the final and the initial fluorometer readings.

Due to time constraints, this elaborate procedure could not be performed on all of the samples. After performing a number of experiments it became apparent that all samples were N and P co-limited. In addition, the difference between filtered and unfiltered samples was relatively insignificant. Based on this information, a much simpler test was designed in order to process as many samples as possible. With this new test, 8 samples were analyzed at one time.

Twenty mL of each sample was added to 6 tubes. Two mL of distilled deionized water was added to 3 of these tubes, while the other 3 received 2 ml of a solution of N and P. One ml of *S. capricornutum* was added to each of the 6 tubes. The same incubation time and conditions as in the more elaborate test were used in this experiment. Only the fluorometer was used for the daily analysis throughout the experiment, as this was the more reliable instrument (Paul Harrison, personal communication)

All glassware used for these experiments were acid washed prior to use. The tests were performed under sterile conditions as much as reasonably possible.

### 3.5 Experimental Methods

This section describes the methodology of the experiments carried out for the vertical and horizontal transects and the containment experiments. A description of the calculations performed with the data from each of these experiments is provided in Appendix 1. The monthly depth weighted average was calculated for each parameter, as outlined in Appendix 1, based on the size of the intervals between the sampling depths.

#### 3.5.1 Primary Production

The  $^{14}\text{C-HCO}_3^-$  incubations were performed to estimate the primary productivity. Samples for radiolabeled bicarbonate ( $^{14}\text{C-HCO}_3^-$ ) incorporation studies were collected in 300 ml BOD bottles, which had been rinsed with sample. Sample from each depth or location to be studied was divided into 3 light bottles and one dark bottle, which served as a control.

One ml of 3.7 uCi/ml  $^{14}\text{C-HCO}_3^-$  was added to each of the BOD bottles, the caps were replaced and the bottles well shaken. These bottles were then incubated for a period of 4 hours, generally starting sometime between 10:00 am and 12:00 noon. Plastic plates, designed to hold the four BOD bottles in a horizontal plane at right angles to one another, were used for this purpose. The BOD bottles were fixed to these plates with the 4 samples from a given depth fixed to the same plate. The plates were then clamped onto a rope and incubated such that samples were suspended at the depth from which they were collected.

$^{14}\text{C}$ -bicarbonate solution (100 ul) was added to duplicate scintillation vials containing 5 ml of scintillation cocktail. These vials were later counted to determine the accurate activity of the solution.

Samples were recovered following four hours of incubation and 1 ml of formaldehyde was added to each bottle. The samples were transported to the UBC Environmental Engineering laboratory for further processing and analysis.

The following procedure was carried out with each sample. The entire 300 ml sample was filtered through a 0.45  $\mu$ m 47 mm cellulose nitrate membrane filter. The sample bottle was washed twice with distilled water and this water was filtered as well. The filter paper was transferred to a labeled scintillation vial and 2-4 drops dilute HCl were added to the vial. The vials were left uncovered overnight and 5 ml of scintillation cocktail (Scintiverse) was added to each one the following morning. Each vial was counted for 5 minutes in a Beckman LS6500 multipurpose scintillation counter operated in an external standard mode to correct for quenching.

Daily primary productivity was estimated based on the total daily solar radiation and the total solar radiation during the incubation. Short wave solar radiation ( $\text{W/m}^2$ ) was recorded daily at 15 minute intervals at a station on the upper basin in Nakusp and at one on the lower basin in Fauquier. Total daily short wave solar radiation on a given sampling date was estimated from an integration of the data from the entire day, while total short wave solar radiation for the incubation period was estimated based on an integration of the data from the incubation period. The total daily primary productivity was estimated by multiplying the total primary productivity for the incubation period by the ratio of the total daily solar radiation to the total solar radiation during the incubation period.

The weather patterns on the incubation date may not be representative of the patterns for the month in which the incubation was performed. Therefore, average daily productivity was estimated for each "month" to dampen the effects of daily variations in the weather. For each basin, the average daily solar radiation totals for each "month" was calculated and this value was used to calculate the average daily primary productivity in the same manner that the actual daily average productivity was calculated. In 1998 the samples were collected towards the end of the month with the average date of collection being the 28<sup>th</sup>. Therefore the average daily productivity was calculated for the period of the 14<sup>th</sup> of one month to the 13<sup>th</sup> of the following month. For example, the productivity for May was calculated using the average daily solar radiation from the period of the 14<sup>th</sup> of May to the 13<sup>th</sup> of June. In general, samples were collected closer to the middle of the month in 1999, with the average sampling date being the 15<sup>th</sup>. Therefore, the average



daily productivity for each month was estimated for the period from the 1<sup>st</sup> day to the last day of the month.

The specific algal growth rate or assimilation rate was estimated for each month by dividing the weighted mean productivity in  $\text{mg C/m}^3/\text{hr}$  by the weighted mean chlorophyll-a in  $\text{mg/m}^3$ . The resulting specific growth rate, which is expressed in  $\text{mg C/mg chlorophyll-a/hour}$  is an indication of the productivity per unit of chlorophyll-a, i.e. the production efficiency, while the primary productivity in  $\text{mg C/m}^3/\text{hr}$  is an indication of the productivity of the entire autotrophic community within the sample.

The depth weighted average hourly primary productivity (units are  $\text{mg C/m}^3/\text{hour}$ ) was calculated, as outlined in Appendix 1, based on the size of the intervals between the sampling depths. The calculations used to estimate daily areal primary productivity (units are  $\text{mg C/m}^2/\text{day}$ ) are also outlined in Appendix 1.

### 3.5.2 Chlorophyll-a

Chlorophyll-a analyses were used to estimate algal biomass. One litre polyethylene bottles were filled with sample from each depth or location and a portion of this sample was used for this procedure. Samples were transported to the UBC Environmental Engineering laboratory in a cooler on ice.

Five hundred mL of sample was filtered through a  $0.45 \mu\text{m}$ , 47 mm cellulose nitrate membrane filter. The filter was placed into the bottom of a petri dish with the side containing filtered material facing up. The petri dishes were each taped shut, wrapped in tin foil and kept in a freezer until further processing.

Each filter paper was transferred into a separate centrifuge tube containing 10 mL of 90% acetone. The tubes were mixed by hand and incubated in the dark at  $4^\circ\text{C}$  for 24 hours. Following the incubation the tubes were centrifuged for 10 minutes at 2000 rpm.

A Turner 10-AU fluorometer was utilized for chlorophyll-a measurements. Sufficient supernatant was decanted from the centrifuge tube into a cuvette and a fluorometer reading was taken. Two drops of 50% HCl were then added to the cuvette and a second reading was taken. These readings were used to calculate the chlorophyll-a concentration in each sample.

### 3.5.3 $^{14}\text{C}$ -Glucose Incorporation

The  $^{14}\text{C}$ -glucose incorporation was used to estimate microbial activity. At each sample depth or location, three sterile 20 mL plastic syringes were rinsed and then filled with 18 mL of sample. Formaldehyde (0.2 mL, 37 %) was added to one of the three syringes from each depth as a control for any physical absorption of the glucose.  $^{14}\text{C}$ -glucose (1 mL of 0.19 uCi/mL, specific activity 170 mCi/mmol) was then added to each syringe, including the formalized controls. Glucose and formaldehyde were added with a 1 mL glass syringe with a metal needle that fit through the small exit orifice of the plastic incubation syringe. The incubation syringes were then fixed to a rope at appropriate positions using an elastic band, such that when the rope was submerged the syringes were positioned at the depth from which the samples originated. A weight was tied to the bottom of the rope to ensure that the syringes were held at the appropriate depths. The syringes were left incubating for 3 hours. They were then recovered and 0.2 mL of formaldehyde was added to the live samples.

The samples were filtered immediately through a 0.2  $\mu\text{m}$  25 mm cellulose nitrate membrane filter in a filter assembly fitted to the syringes. The filter was washed 2 to 3 times with lake water. The filter was then placed into a scintillation vial containing 5 mL of scintillation cocktail. The vials were transported to the UBC environmental engineering laboratory and analyzed using the scintillation counter operated in an external standard mode to correct for quenching.

$^{14}\text{C}$ -glucose solution (100  $\mu\text{L}$ ) used in this experiment was added in duplicate to scintillation vials containing 5 mL of scintillation solution. These vials were counted to determine the exact activity of the solution used in the experiment.

### 3.5.4 DAPI

The DAPI method was performed to estimate bacterial abundance (biomass). The following procedure, based on Porter and Feig (1980), was used to measure the abundance of bacteria in samples. Ten mL of sample was added to a capped 20 mL sterile glass test tube. Formaldehyde (0.20 mL) was added to each tube and they were kept in a cooler on ice. The tubes were transported to the UBC environmental

engineering laboratory where they remained in a cold room at 4 °C until further processing and analysis.

The following procedure was carried out for each sample. DAPI (4', 6-diaminidino-2-phenylindole) (200 uL of a 10 mg/L solution) was added to a tube and this sample was incubated in the dark for 5 minutes. The sample was filtered through a 0.20 um 25 mm black Nucleopore polycarbonate filter. The filtration apparatus was set up such that the polycarbonate filter was mounted onto a 25 mm glass fiber filter in order to ensure an even distribution of bacteria on the filter.

Following filtration, the filter was mounted over a drop of immersion oil on a slide. A drop of immersion oil was then placed on top of the filter, followed by a cover slip. The slide was observed under the 100X objective of a Zeiss epifluorescence microscope, using the #1 adsorption filter, and a 10 x 10 grid mounted in the left objective was used for counting bacteria. The number of bacteria were counted in as many squares as possible before the fluorescence began to fade. The number of bacteria per number of squares was recorded and the procedure was repeated 8 to 12 times.

### **3.5.5 CTC and INT**

The CTC and INT methods were used to estimate the percentage of active bacteria. Ten mL of sample from each depth was added to individual capped 20 mL glass test tubes. These samples were transported to the UBC Environmental Engineering Laboratory on ice and then transferred to a 4 °C cold room until further processing.

Initially, the INT (2-(p-iodophenyl)-3-(p-nitrophenyl)-5-phenyl tertazolium chloride) method was used to estimate the percentage of active bacteria. However, no active bacteria were detected using this method. Therefore the CTC (5-cyano-2,3-ditolyl tetrazolium chloride) method was used in its place starting in July 1998.

The following procedure, modified from Rodriguez et al. (1992), was carried out when the CTC method was used. One ml of 7.7 g/L CTC was added to each tube and they were incubated in the dark at 4 °C for approximately 24 hours. Formaldehyde (0.1 mL, 37 %) was added to each tube to terminate the incubation. Each sample was filtered through a black 0.2 um 25 mm polycarbonate filter. As with the DAPI procedure, the filtration apparatus was set up such that the polycarbonate filter was mounted onto a 25

mm glass fiber filter in order to ensure an even distribution of bacteria on the filter. Following filtration, the filter was mounted on a slide and cells were counted in the same manner as for the DAPI procedure. The #15 adsorption filter was used for this procedure.

When the INT procedure was performed the following procedure, a modification of Zimmermann et al. (1978) was carried out. One ml of a 0.2% aqueous INT solution was added to each tube and the tubes were incubated for 20 minutes in the dark at 4 °C. Formaldehyde (0.1 mL, 37%) was added to each tube to terminate the incubation. Five mL of each sample was then filtered through a black 0.2  $\mu$ m 25 mm Nucleopore polycarbonate filter mounted on the filtration apparatus in the same manner as in the CTC procedure. Following filtration, 1 mL of an acridine orange solution (1:10 000 in 6.6 mM phosphate buffer, pH 6.7) was added on top of the filter and left to stain for 2 minutes. This solution was then removed by filtration and the filter was partially air dried for a few minutes. The filter was mounted on a slide and cells were counted in the same manner as for the DAPI procedure. The #9 adsorption filter was used for this procedure.

### **3.5.6 Alkalinity**

The alkalinity measurements were performed according to the method outlined in Standard Methods (APHA 1995). One hundred ml of sample, from the 1 L polyethylene bottle, was transferred into a 250 ml erlenmeyer flask and the pH (Beckman 44) was measured. This sample was then titrated with 0.02 N H<sub>2</sub>SO<sub>4</sub> to a pH of 4.5. All samples had an initial pH of less than 8.3. Therefore, the lake water had no P alkalinity and the M.O. alkalinity was equal to the total alkalinity. Titrations were performed in duplicate or triplicate to check the precision of the results.

This procedure was carried out with the samples from 0 m and 15 m from each lake. In addition, when the alkalinities of these samples varied significantly, alkalinity titrations were performed with samples from at least one other depth.

### **3.6 Statistical analyses**

The JMP Start Statistics software package was utilized for all statistical analyses. This section describes the types of analyses performed for each type of experiment.

### 3.6.1 Vertical transects

Individual statistical analyses were performed with the data from the incubations, chlorophyll-a analyses and DAPI experiments. Using the data from a given experiment, a general linear model was run with a full factorial of the following effects: location, year and month, where location is the basin, and year and month indicate when the sample was collected. The following contrasts were performed with respect to the location \* year effect to determine whether differences from one basin to the other and from one year to the next were significant at the 95 % confidence level ( $p \leq 0.05$ ):

- lower basin 1998 vs. lower basin 1999
- upper basin 1998 vs. upper basin 1999
- lower basin 1998 vs. upper basin 1998
- lower basin 1999 vs. upper basin 1999

### 3.6.2 Horizontal transects

Statistical analyses of the data from the chlorophyll-a analyses and  $^{14}\text{C-HCO}_3^-$  incorporation experiment were performed. Using the data from a given experiment, a general linear model was run with the month as an effect. The following contrasts with respect to this effect were performed:

- June vs. August
- June vs. September
- August vs. September

These contrasts were performed to determine whether the differences in each parameter from month to month along a transect of the lake were significant at the 95 % confidence level.

### 3.6.3 Containment Experiments

A grouped means one way ANOVA was performed with the primary productivity data from the containment experiments. The significance of the difference between the means of the primary productivity of the four carboys was assessed at the 95 % confidence level ( $p \leq 0.05$ ).

### 3.6.4 Stream bioassays

Statistical analyses were performed on the data from each bioassay to determine whether the addition of one nutrient or a combination of nutrients resulted in a significant increase in growth. For a given bioassay, the net growth was calculated for each test tube and a statistical model was created using this data. In each case, the model was run with treatment, filter and treatment \* filter as effects, where treatment is the nutrient or combination of nutrients added to the test tube and filter is an indication of whether or not the sample was filtered. As previously described in the stream bioassay section, the following treatments were used in the bioassays: N; N+Nu; P; P+Nu; Nu; N+P; N+P+Nu; and distilled, deionized water (control), where N = nitrogen, P = phosphorus and Nu = micronutrients. Each of these treatments were used on filtered and unfiltered water. The following contrasts of the different treatments were then performed:

- filtered vs unfiltered (f vs. nf);
- all samples with N vs. all samples with P (N vs. P);
- all samples with a combination of N and P vs. all other samples (N+P vs. others)
- control vs. all others except those with a combination of N and P (Ctrl vs. others);
- N, N+Nu vs. P, P+Nu, control, micronutrients (N vs. P & Ctrl & Nu)
- P, P+Nu vs. N, N+Nu, control, micronutrients (P vs. N & Ctrl & Nu)
- presence of micronutrients vs. absence of micronutrients (Nu vs w/o Nu)

## 4. RESULTS

### 4.1 Vertical Transects

#### 4.1.1 Primary Productivity

The results from each basin (upper and lower) during each year (1998, the pre-treatment year, and 1999, the first year of fertilization) will be discussed individually. A general discussion will then follow. The monthly maximum and minimum primary productivity in the upper basin, and the depths at which they were measured, are presented in Table 4.1, while those in the lower basin are presented in Table 4.2. Also included in these tables are the areal primary productivity and the estimated monthly average areal primary productivity (expressed as a daily rate) as well as the light intensity at the surface and the compensation depths.

##### Upper Basin 1998

The monthly primary productivity profiles in the upper basin of the Arrow Reservoir in 1998 are depicted in Figure 4.1. In May and September, the primary productivity was relatively low, peaking at 0.85 and 0.68 mg C/m<sup>3</sup>/hr, respectively, at a depth of 5 m. The productivity was slightly greater in June, July and August when bimodal productivity profiles were observed. The first peak was at a depth of 1 m (1.69 mg C/m<sup>3</sup>/hr in June, 2.09 mg C/m<sup>3</sup>/hr in July and 1.86 mg C/m<sup>3</sup>/hr in August), while the second smaller peak was at 5 m in June, 5 to 10 m in July and 10 m in August. This bimodal productivity profile was not observed in this basin in 1999.

The light intensity readings at the surface were relatively high in July, August and September, at 1050, 950 and 1025 uEinst./m<sup>2</sup>/s, respectively, but in May and June, the values were slightly lower, 865 and 800 uEinst./m<sup>2</sup>/s. The compensation depths were 15 m in May, 16 m in June, 17.5 m in July, 17 m in August and 18 m in September.

##### Lower Basin 1998

The monthly primary productivity profiles in the lower basin of the Arrow Reservoir in 1998 are depicted in Figure 4.1. As with the upper basin, the primary

Table 4.1 - Summary of primary productivity results for the upper basin of the Arrow Reservoir

| Year<br>Month                     | 1998 |       |       |        |       | 1999  |       |       |       |        |       |
|-----------------------------------|------|-------|-------|--------|-------|-------|-------|-------|-------|--------|-------|
|                                   | May  | June  | July  | August | Sept. | April | May   | June  | July  | August | Sept. |
| Maximum (mg C/m <sup>3</sup> /hr) | 0.85 | 1.69  | 2.09  | 1.86   | 0.68  | 1.55  | 4.38  | 1.57  | 8.3   | 7.9    | 0.50  |
| Depth (m)*                        | 5    | 1     | 1     | 1      | 5     | 10    | 2     | 2     | 1     | 1      | 2     |
| Minimum (mg C/m <sup>3</sup> /hr) | 0.05 | 0.24  | 0.87  | 0.75   | 0.10  | 0.28  | 0.48  | 0.02  | 0.07  | 0.40   | 0.08  |
| Depth (m)**                       | 0    | 15    | 0     | 15     | 0     | 0     | 15    | 15    | 15    | 15     | 15    |
| Areal (mg C/m <sup>2</sup> /hr)   | 8.23 | 13.75 | 25.66 | 22.25  | 6.91  | 18.07 | 29.20 | 7.80  | 39.00 | 44.82  | 3.90  |
| Monthly avg. areal                | 40.2 | 115.5 | 245.7 | 175.9  | 55.2  | 143.9 | 598.1 | 114.3 | 250.7 | 254.4  | 23.3  |
| Surface light intensity           | 865  | 800   | 1050  | 950    | 1025  | 510   | 1000  | 375   | 1200  | 1100   | 885   |
| Compensation depth (m)            | 15   | 15    | 17.5  | 17     | 18    | >26   | 24    | 7.5   | 10.5  | 11     | 15    |

Monthly average areal primary productivity units are mg C/m<sup>2</sup>/day, surface light intensity units are uEinsteins/m<sup>2</sup>/s

\* Depth of maximum productivity

\*\* Depth of minimum productivity

\*\*\* Weighted average hourly primary productivity

Table 4.2 - Summary of primary productivity results for the lower basin of the Arrow Reservoir

| Year<br>Month                     | 1998 |       |       |        |       | 1999  |      |      |       |        |       |
|-----------------------------------|------|-------|-------|--------|-------|-------|------|------|-------|--------|-------|
|                                   | May  | June  | July  | August | Sept. | April | May  | June | July  | August | Sept. |
| Maximum (mg C/m <sup>3</sup> /hr) | 0.54 | 1.08  | 3.8   | -      | 0.67  | 2.49  | 0.2  | -    | 2.24  | -      | 0.57  |
| Depth (m)*                        | 5    | 5     | 10    | -      | 2     | 10    | 10   | -    | 2     | -      | 5     |
| Minimum (mg C/m <sup>3</sup> /hr) | 0.11 | 0.27  | 0.27  | -      | 0.32  | 0.45  | 0.05 | -    | 0.30  | -      | 0.15  |
| Depth (m)**                       | 15   | 15    | 0     | -      | 10    | 0     | 0    | -    | 15    | -      | 15    |
| Areal (mg C/m <sup>2</sup> /hr)   | 5.45 | 11.87 | 43.73 | -      | 6.88  | 28.86 | 2.65 | -    | 21.82 | -      | 6.42  |
| Monthly avg areal                 | 27.0 | 76.3  | 361.6 | -      | 56.0  | 295.5 | 31.7 | -    | 142.9 | -      | 48.5  |
| Surface light intensity           | 435  | 1200  | 1100  | 1050   | 1075  | 480   | 550  | -    | 1150  | -      | 940   |
| Compensation depth (m)            | 16   | 16    | 18.5  | 24     | 22    | 18.5  | >25  | -    | 14.5  | -      | 16    |

\*.- No samples collected during this period.

Monthly average areal primary productivity units are mg C/m<sup>2</sup>/day, surface light intensity units are uEinsteins/m<sup>2</sup>/s

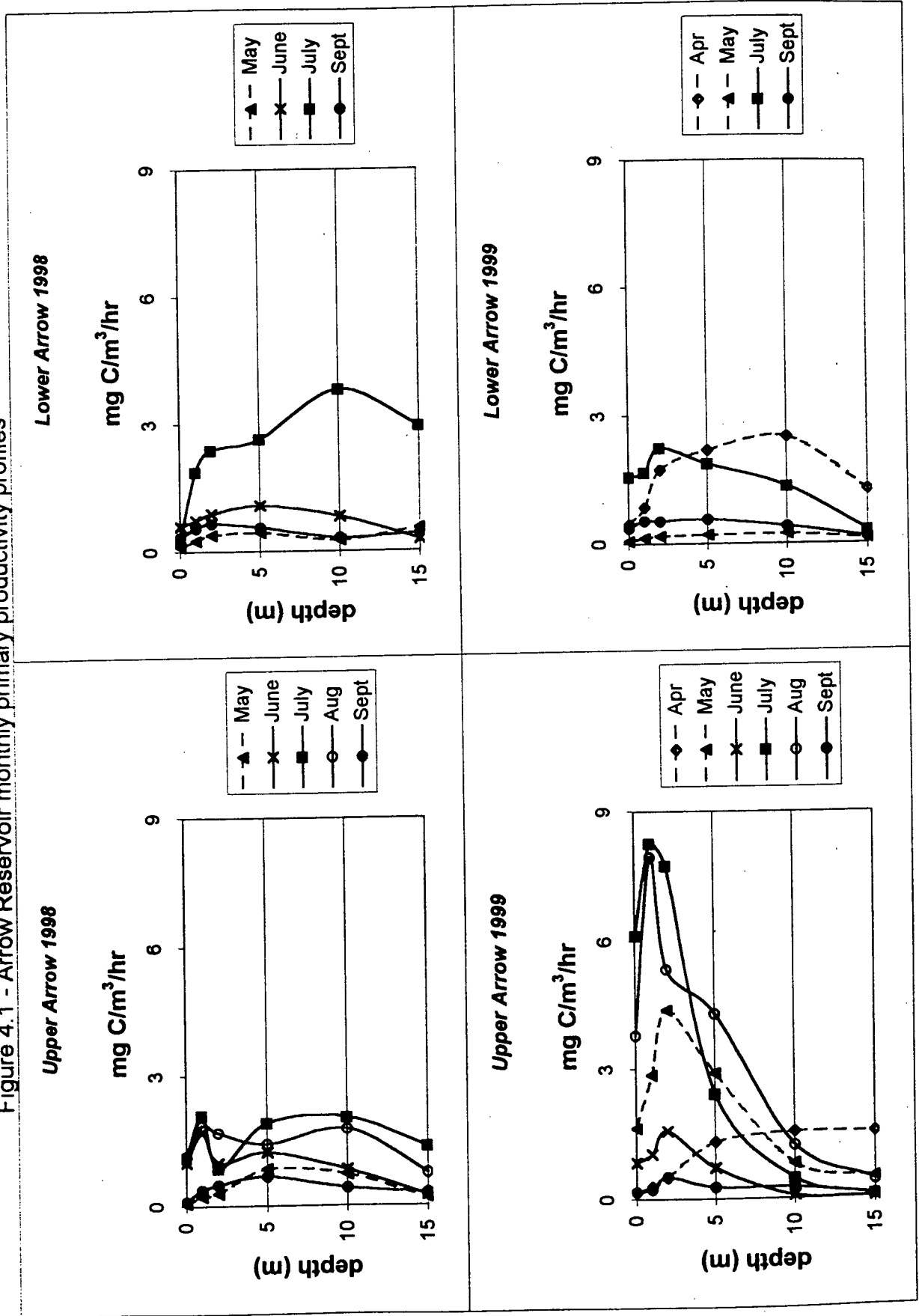
\* Depth of maximum productivity

\*\* Depth of minimum productivity

\*\*\*Weighted average hourly primary productivity



Figure 4.1 - Arrow Reservoir monthly primary productivity profiles



productivity in the lower basin was relatively low in May and September. During these months, a bimodal productivity profile was observed with peaks at 2 to 5 m ( $0.4 \text{ mg C/m}^3/\text{hr}$ ) and 15 m ( $0.54 \text{ mg C/m}^3/\text{hr}$ ) in May and at 2 m ( $0.67 \text{ mg C/m}^3/\text{hr}$ ) and 15 m ( $0.41 \text{ mg C/m}^3/\text{hr}$ ) in September. The primary productivity was only slightly greater in June and substantially greater in July. A single peak in primary productivity was recorded during these summer months:  $1.08 \text{ mg C/m}^3/\text{hr}$  at 5 m in June and  $3.80 \text{ mg C/m}^3/\text{hr}$  at 10 m in July.

The light intensity at the surface was greater than  $1000 \text{ uEinst./m}^2/\text{s}$  from June until September. In May the light intensity at the surface was quite low at only  $435 \text{ uEinst./m}^2/\text{s}$ . The compensation depths were at approximately 16.5 m in May, 16 m in June, 18.5 m in July and 21.5 m in September.

#### Upper Basin 1999

The monthly primary productivity profiles in the upper basin of the Arrow Reservoir in 1999 are presented in Figure 4.1. The primary productivity peaked close to the surface during each month of the sampling season, with the exception of April (It is worth noting here that fertilization began less than a week after the April samples were collected). The productivity in April, which was very low at the surface ( $0.16 \text{ mg C/m}^3/\text{hr}$ ), exhibited a broad peak of approximately  $1.5 \text{ mg C/m}^3/\text{hr}$  from 10 to 15 m. The highest peaks in this basin in 1999 were  $8.26$  and  $7.94 \text{ mg C/m}^3/\text{hr}$ , observed in July and August, respectively, at 1 m. These peaks were substantially greater than any others recorded in the reservoir during this study. The peaks in May, June and September were all observed at a depth of 2 m. Of these three months, May had the highest peak at  $4.38 \text{ mg C/m}^3/\text{hr}$ , followed by June at  $1.57 \text{ mg C/m}^3/\text{hr}$  and September at  $0.50 \text{ mg C/m}^3/\text{hr}$ .

The light intensity at the surface was low in April ( $510 \text{ uEinst./m}^2/\text{s}$ ) and June ( $375 \text{ uEinst./m}^2/\text{s}$ ). This parameter was relatively high in May ( $1000 \text{ uEinst./m}^2/\text{s}$ ), July ( $1200 \text{ uEinst./m}^2/\text{s}$ ) and August ( $1100 \text{ uEinst./m}^2/\text{s}$ ) and moderate in September ( $885 \text{ uEinst./m}^2/\text{s}$ ). The light penetration was relatively deep in the spring with compensation depths of below 25 m in April and approximately 23 m in May. The light penetration in the summer was relatively shallow with compensation depths of approximately 7.5 m in June, 10.5 m in July and 11 m in August.

### Lower Basin 1999

The monthly primary productivity profiles in the lower basin of the Arrow Reservoir in 1999 are presented in Figure 4.1. The greatest primary productivity in this basin was recorded in April, while the lowest was observed in September. Primary productivity declined from April to May, increased from May to July and then declined to September. During the spring, deeper, broader primary productivity peaks were observed with maximum values of 2.49 and 1.23 mg C/m<sup>3</sup>/hr at a depth of 10 m in April and May, respectively. In mid-summer, a more distinct peak was recorded at 2 m (2.24 mg C/m<sup>3</sup>/hr), while the maximum productivity in September was observed from 1 to 5 m (0.54 to 0.57 mg C/m<sup>3</sup>/hr)

The light intensity was relatively low in the spring with surface values of 480 and 550 uEinst./m<sup>2</sup>/s recorded in April and May, respectively. In July, a relatively high light intensity of 1150 uEinst./m<sup>2</sup>/s was recorded at the surface and it remained moderately high in September (940 uEinst./m<sup>2</sup>/s). Light penetration was greatest in the spring with compensation depths of 18.5 m in April and greater than 25 m in May. In July and September the compensation depths were 14.5 and 16 m, respectively.

### General

The results of the statistical analysis indicate that the primary productivity of the upper basin is significantly greater in 1999 than in 1998 (p value = 0.008), while there is no significant difference in the primary productivity in the lower basin between 1998 and 1999 (p value = 0.8). In addition, in 1999, the primary productivity in the upper basin was significantly greater than that in the lower basin (p value = 0.01). In 1998, there was no significant difference in the productivity of the two basins (p value = 0.7).

The seasonal areal primary productivity and daily areal primary productivity are depicted in Figures 4.2 and 4.3a, respectively. These data show a peak in primary productivity occurred during the summer in each basin during both years of the study. A spring peak was also noted in both basins in 1999. Primary productivity in the upper basin was greater during the first year of fertilization in 1999 than during the year prior to fertilization in 1998. While there was no statistical difference between the annual

Figure 4.2 Arrow Reservoir Areal Primary Productivity Seasonal Profiles

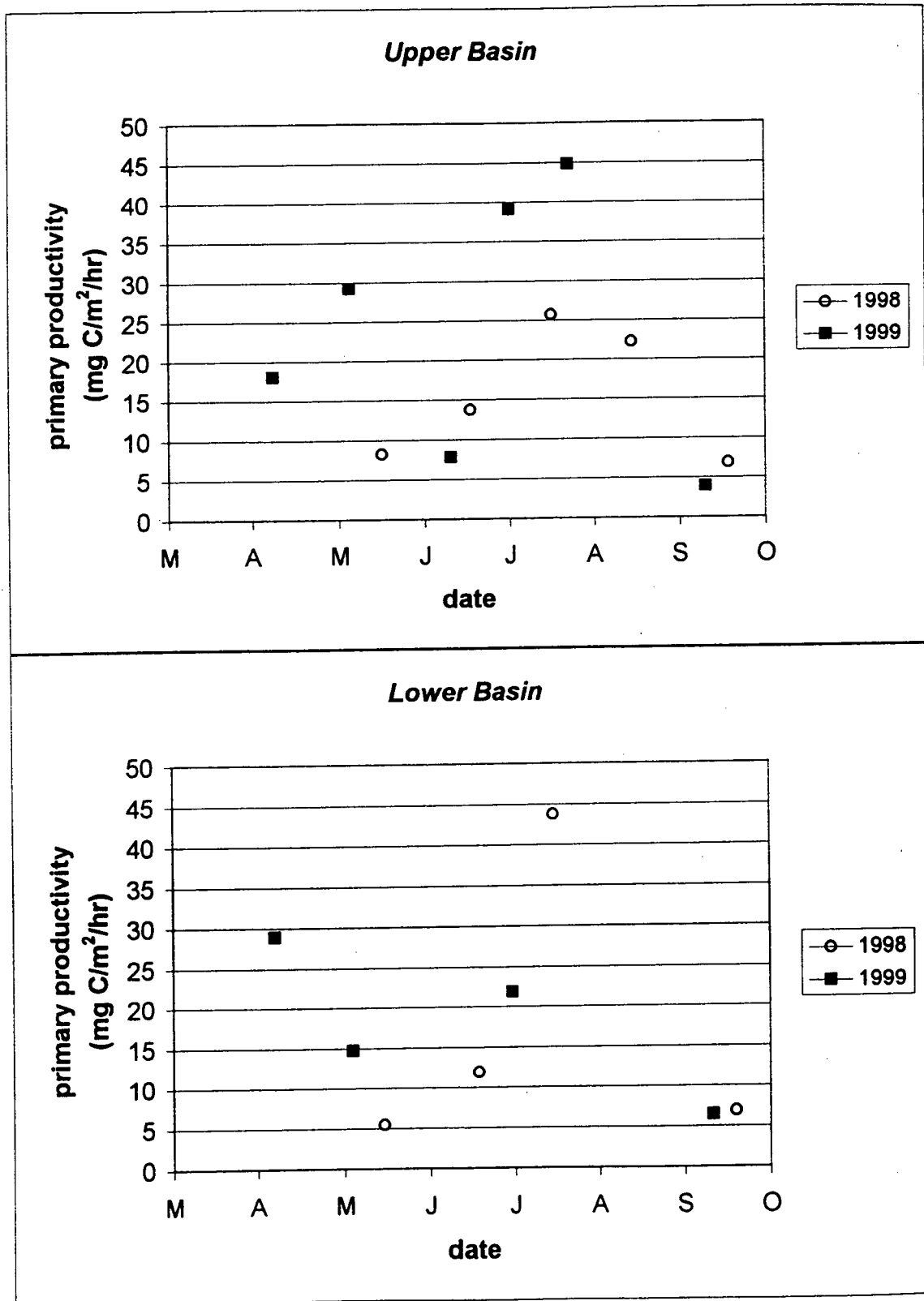
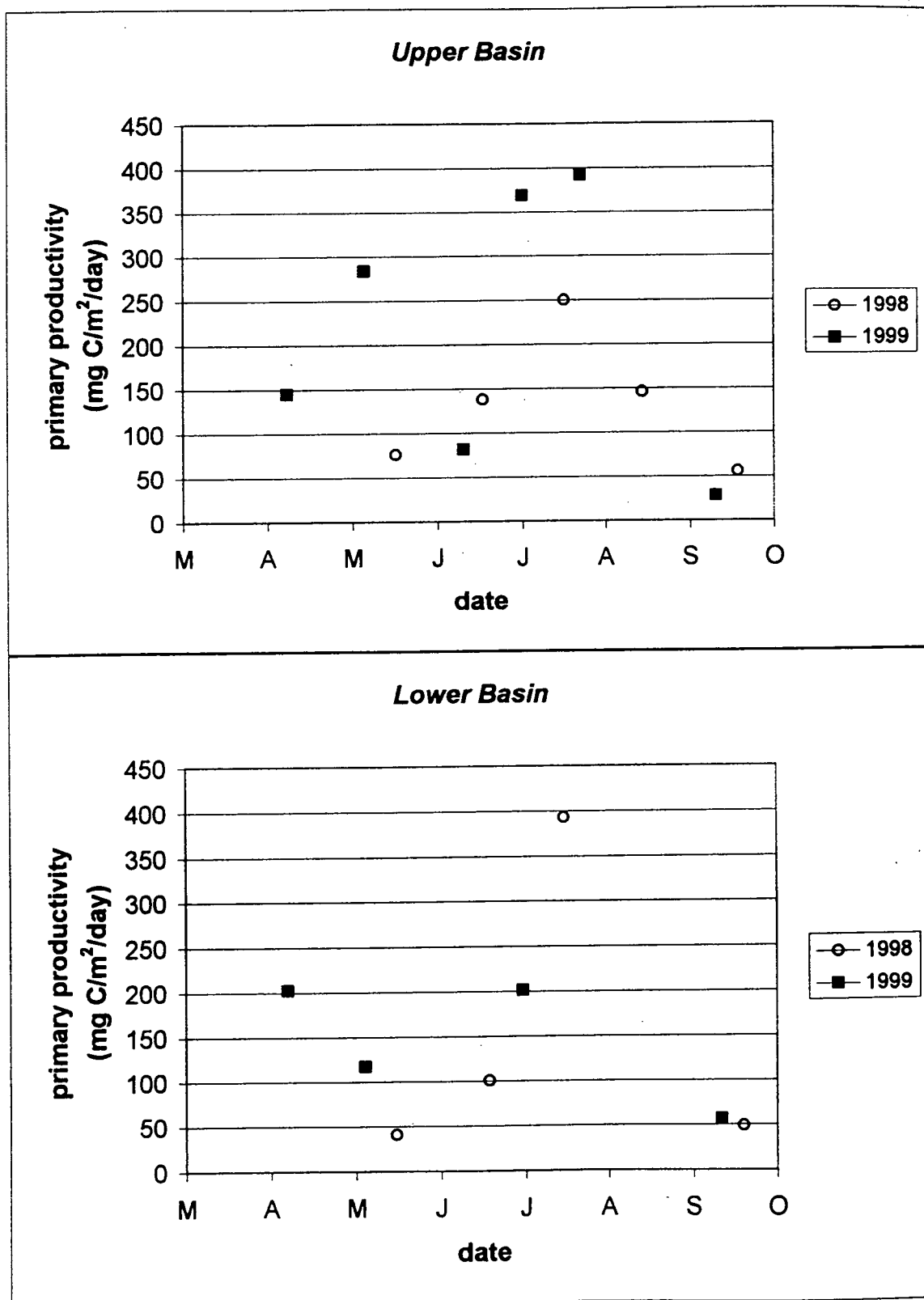


Figure 4.3a Arrow Reservoir daily areal primary productivity seasonal profiles



primary productivity in the lower basin in 1998 and 1999, it appears that this parameter may have been greater in 1998 than 1999 in mid-summer.

The weather conditions on a given sampling day may not be representative of those for the period when the samples were collected. In order to take into account the daily variations in weather conditions, the monthly average productivity (expressed as a daily rate) was calculated for a period of approximately one month surrounding the sampling date using the average daily solar radiation for that period. The seasonal variation in monthly average areal productivity (expressed as a daily rate) is depicted in Figure 4.3b. It should be noted that for 1998 (when samples were collected at the end of the month), "month" indicates the period of approximately 15 days prior to and 15 days following the sampling date, while for 1999 (when samples were collected mid-month) "month" indicates the period from the first day to the last day in the month during which the sample was collected.

During 1998, there was very little difference between the daily areal primary productivity and the monthly average areal productivity (see Figures 4.3a and 4.3b), indicating that the daily areal primary productivity is representative of the period in which it was measured. In 1999, the trends in daily areal primary productivity and monthly average areal primary productivity were similar. However, while the largest peak in daily areal primary productivity was recorded in the summer, the greatest monthly average areal primary productivity was observed in the spring. This is due to the fact that, in 1999, spring samples were collected on days with relatively low solar radiation while summer samples were collected on days with relatively high solar radiation.

The trends in specific growth rate (shown in Figure 4.4) were similar to those for primary productivity (shown in Figures 4.2, 4.3a and 4.3b) throughout the study. In 1998, a single peak in both parameters was observed in July. During the following year, two peaks were observed in each basin. In the lower basin, both primary productivity and specific algal growth rate peaked in April and again in July. In the upper basin, the first peak in primary productivity was observed in May and the second in July and August with the maximum productivity occurring in August. The specific algal growth

Figure 4.3b Arrow Reservoir monthly average areal primary productivity profiles

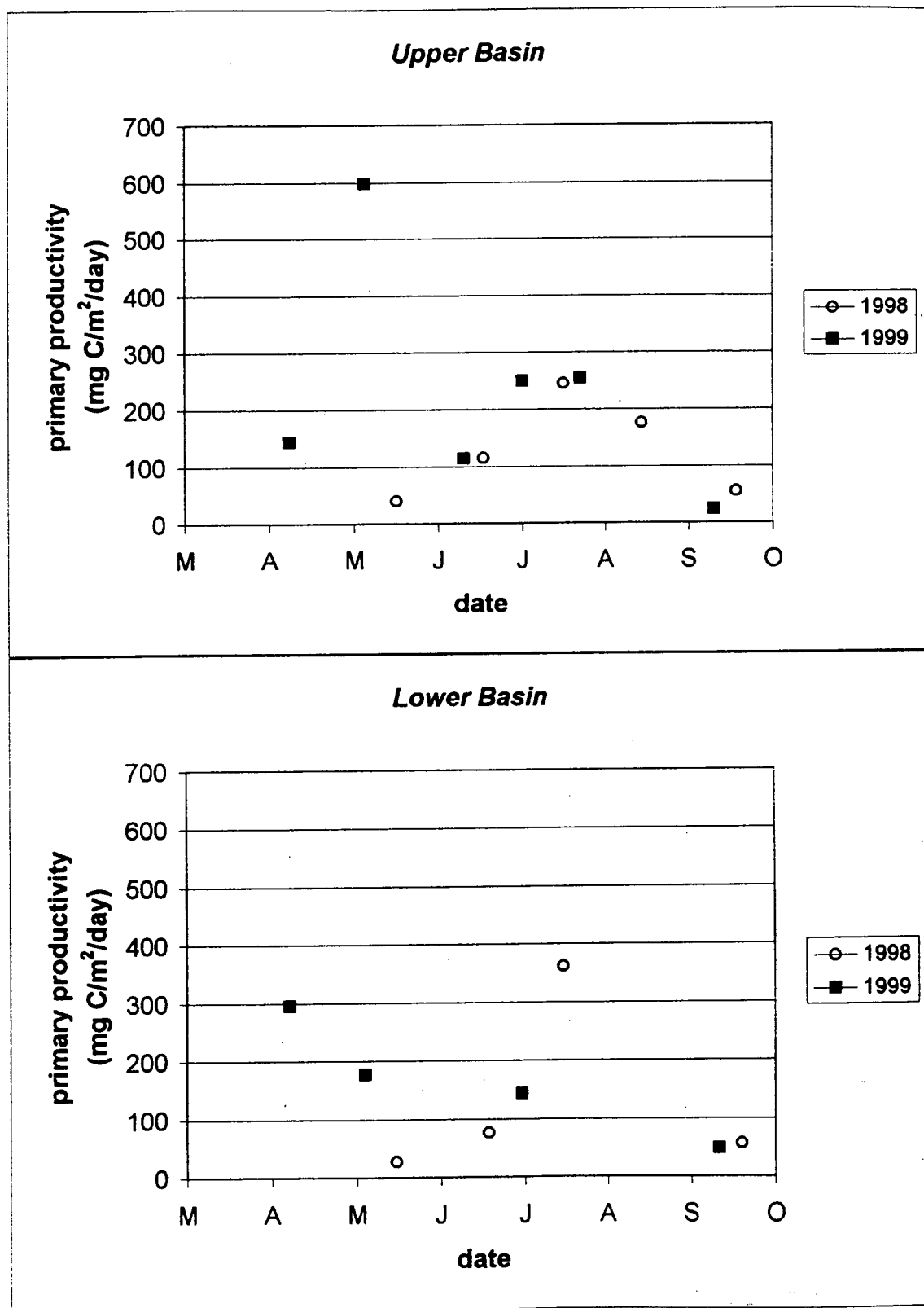
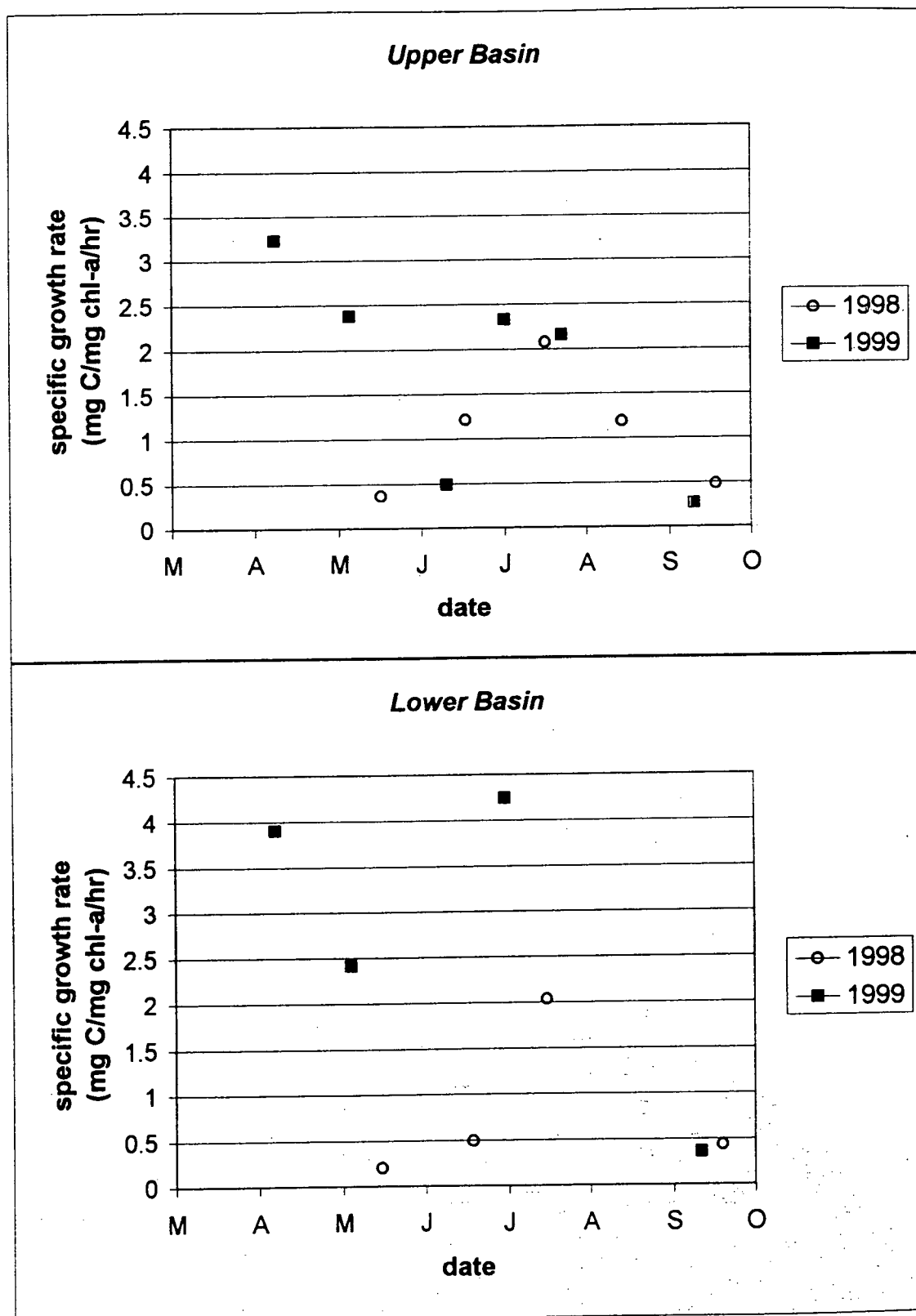


Figure 4.4 Arrow Reservoir specific growth rate seasonal profile





rate in this basin peaked first in April and May (with the maximum value observed in May) and then again in July.

#### 4.1.2 Chlorophyll-a

The chlorophyll-a results for each basin are discussed separately. A general discussion of the treatment of the data and the statistical analyses follows. The monthly depth weighted average chlorophyll-a concentrations in the lower and upper basins are summarized in Table 4.3.

Table 4.3 Summary of the Arrow Reservoir chlorophyll-a results

| Year | Basin | Depth weighted average chlorophyll-a (mg/m <sup>3</sup> ) |      |      |      |      |       |
|------|-------|---|------|------|------|------|-------|
|      |       | April   | May  | June | July | Aug. | Sept. |
| 1999 | Upper | 0.37  | 0.82 | 1.07 | 1.12 | 1.39 | 0.98  |
|      | Lower | 0.49  | 0.41 | -    | 0.34 | 1.27 | 1.20  |
| 1998 | Upper | -   | 1.51 | 0.76 | 0.82 | 1.25 | 0.96  |
|      | Lower | -   | 1.78 | 1.61 | 1.44 | 1.36 | 1.05  |

"-" No samples collected during this period

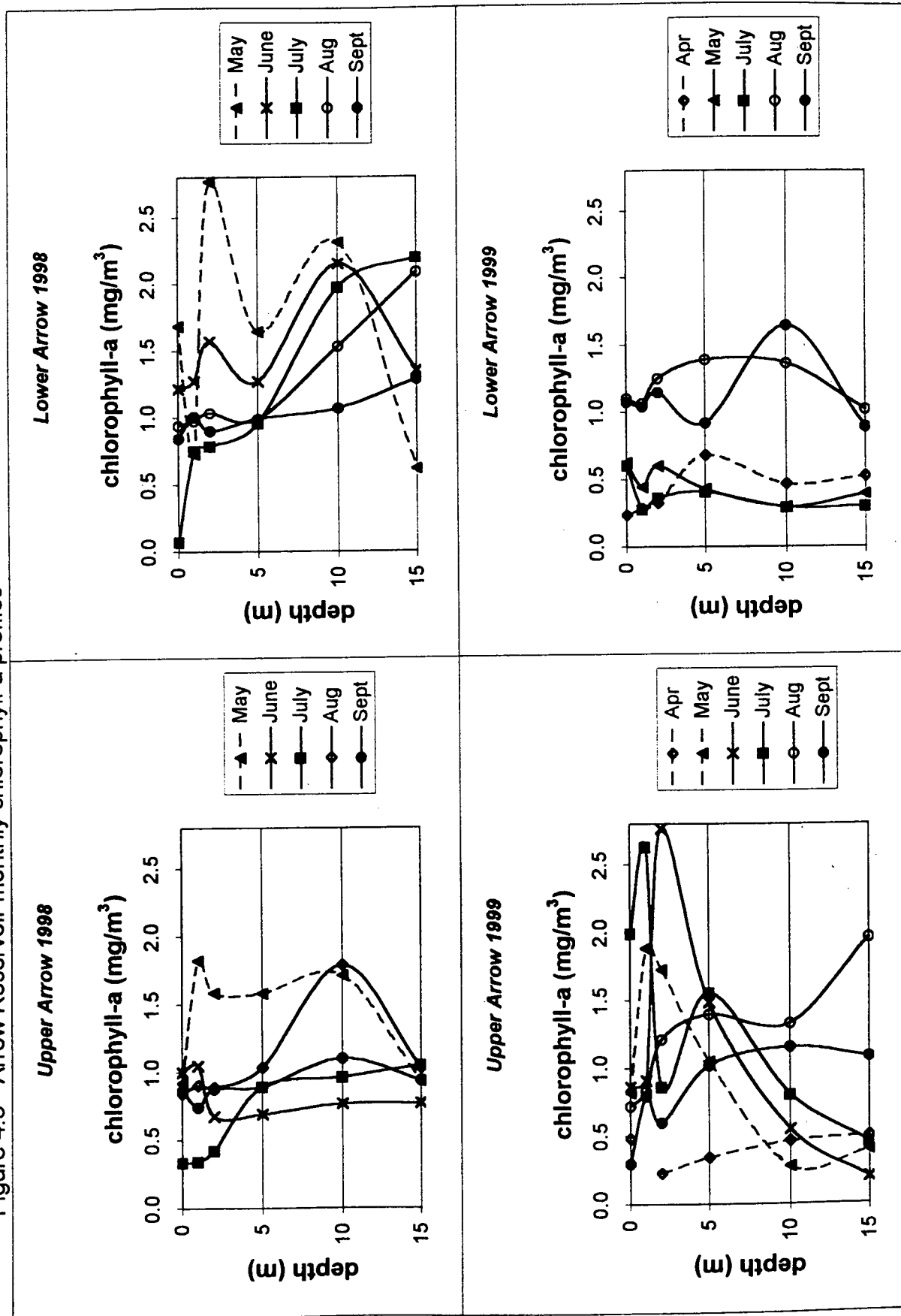
#### Upper Basin 1998

The monthly chlorophyll-a profiles in the upper basin in 1998 are depicted in Figure 4.5. These profiles were variable from month to month and there were no variations of greater than 3X in this parameter with depth or with time. Chlorophyll-a was greatest in May and lowest in June and July. There was a small increase in chlorophyll-a in August followed by a decrease in September.

#### Lower Basin 1998

In 1998, there was a gradual decline in chlorophyll-a in the lower basin from May until September. As with the upper basin, the profiles were variable from month to month. The monthly chlorophyll-a profiles in the lower basin are depicted in Figure 4.5. Near the surface, chlorophyll-a was greater early in the season, while, at 15 m, chlorophyll-a was lowest in May, peaking in July and August.

Figure 4.5 - Arrow Reservoir monthly chlorophyll-a profiles



### Upper Basin 1999

The monthly chlorophyll-a profiles in the upper basin in 1999 are depicted in Figure 4.5. Chlorophyll-a increased consistently throughout the season in the upper basin in 1999, peaking in August and declining slightly in September. In April, chlorophyll-a was quite low, exhibiting little variation with depth. A distinct peak in chlorophyll-a was observed near the surface in May, June and July. In contrast, this parameter was greatest in the deepest samples (those collected from 10 and 15 m) near the end of the season, in August and September.

### Lower Basin 1999

In 1999, chlorophyll-a was relatively low in the lower basin from April until July, exhibiting little variation with depth. It should be noted that no samples were collected in June. Chlorophyll-a was substantially greater in August when it was double that of the previous months, although there was still little variation with depth. The greatest values of chlorophyll-a in the lower basin in 1999 were measured in September, when a peak of  $1.65 \text{ mg/m}^3$  was observed at 10 m. The monthly chlorophyll-a profiles in the lower basin in 1999 are depicted in Figure 4.5.

### General

The results of the statistical analyses indicate that, in the lower basin, the chlorophyll-a in 1998 was statistically greater than that in 1999 ( $p = 0.02$ ). There was no significant difference between the two years in the upper basin ( $p = 0.2$ ). A comparison of the chlorophyll-a between basins revealed that there was no significant difference in 1998 ( $p = 0.1$ ), while the concentration in the upper basin was significantly greater than in the lower basin in 1999 ( $p = 0.04$ ). The seasonal trends in phytoplankton biomass (chlorophyll-a) are depicted in Figure 4.6.

#### **4.1.3 $^{14}\text{C}$ -glucose incorporation**

The monthly bacterial activity profiles in the upper and lower basins in 1998 and 1999 are presented in Figure 4.7. The  $^{14}\text{C}$ -glucose incorporation rate was undetectable in

Figure 4.6 Seasonal profiles of depth weighted average phytoplankton abundance in the Arrow Reservoir

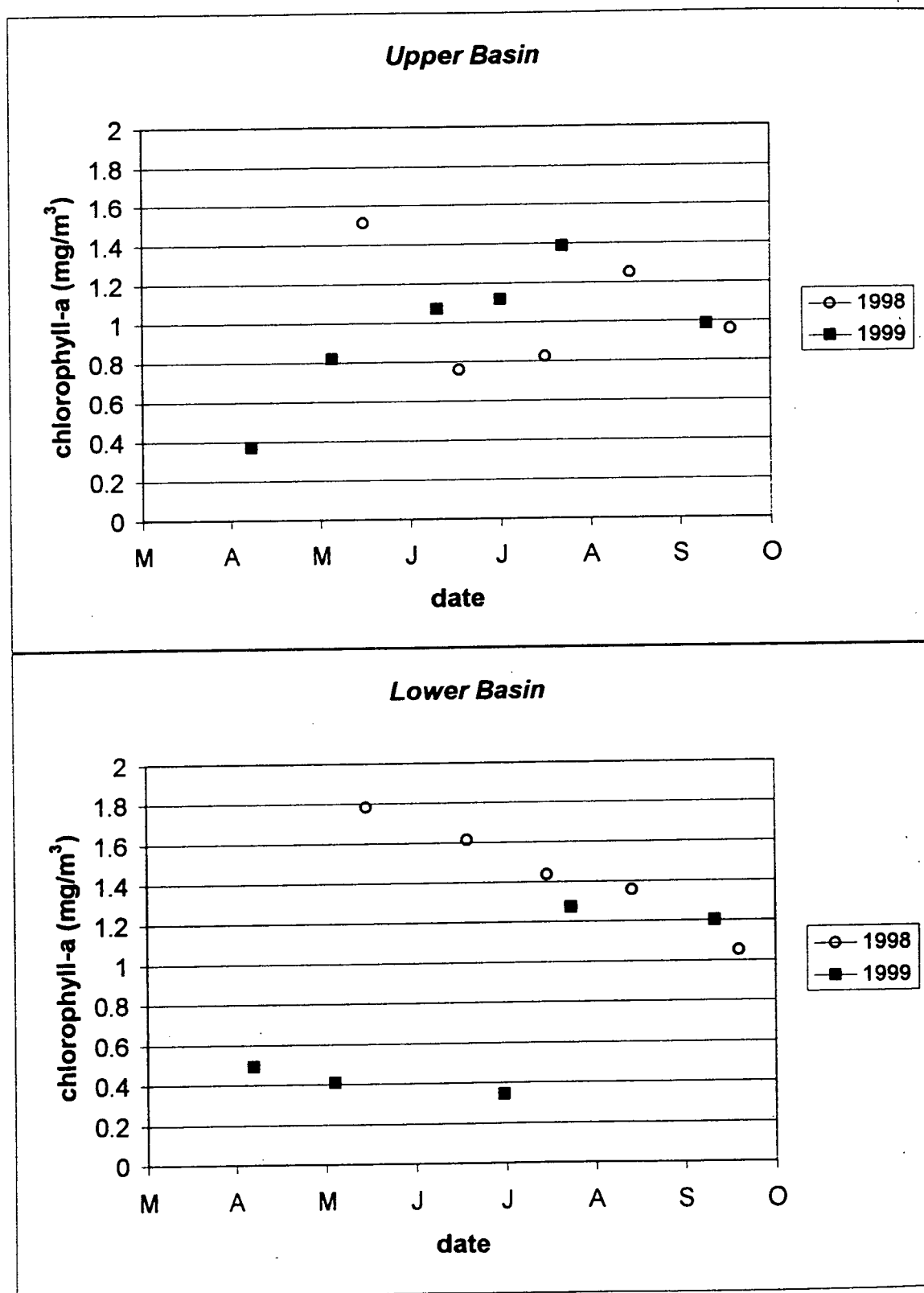
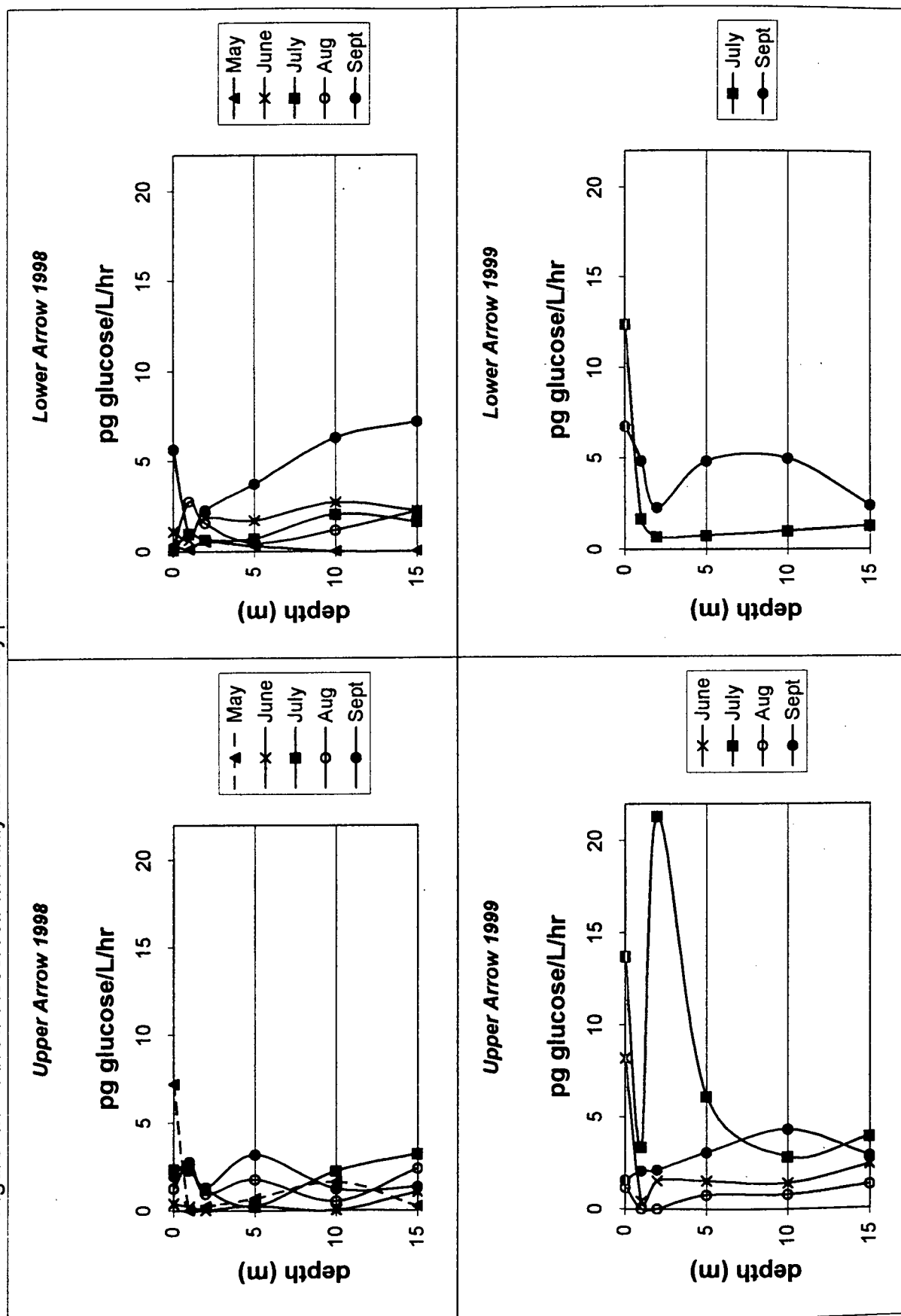


Figure 4.7 - Arrow Reservoir monthly bacterial activity profiles



both basins of the reservoir in April and May of 1999. The monthly depth weighted average bacterial activity in the upper and lower basins of the reservoir in 1998 and 1999 are presented in Table 4.4.

Table 4.4 Summary of the Arrow Reservoir bacterial activity results

| Year | Basin | Depth weighted average bacterial activity (pg glucose/L/hr) |      |      |      |      |       |
|------|-------|---|------|------|------|------|-------|
|      |       | April   | May  | June | July | Aug. | Sept. |
| 1999 | Upper | NR  | NR   | 2.56 | 8.53 | 0.66 | 2.66  |
|      | Lower | NR  | NR   | -    | 2.96 | -    | 4.34  |
| 1998 | Upper | -   | 1.65 | 0.32 | 1.92 | 1.58 | 1.96  |
|      | lower | -   | 0.21 | 1.70 | 1.02 | 1.39 | 4.35  |

"-" No samples collected during this period

NR – No results obtained during this period

There was no significant difference in the microbial activity in the two basins in 1998 (p value = 0.99) or 1999 (p value = 0.2). In addition, the difference in this parameter in the lower basin in 1998 and 1999 is not significant (p value = 0.5). In the upper basin, the microbial activity in 1999 was significantly greater than that in 1998 (p value = 0.05).

A profile of weighted average bacterial activity as a function of month is presented in Figure 4.8. This figure shows that, in general, there was an increase in the bacterial activity throughout the season. There was, however, a relatively large peak in this parameter in the upper basin in July 1999. In the lower basin, where data is available, the difference between the bacterial productivity in 1998 and 1999 is minimal. In the upper basin, with the exception of the large peak in July 1999, the annual differences are relatively small.

#### 4.1.4 DAPI

The monthly bacterial abundance profiles in the upper and lower basins in 1998 and 1999 are presented in Figure 4.9. The monthly depth weighted average bacterial abundance in each basin in 1998 and 1999 is presented in Table 4.5.

Figure 4.8 Seasonal bacterial activity profiles for the Arrow Reservoir

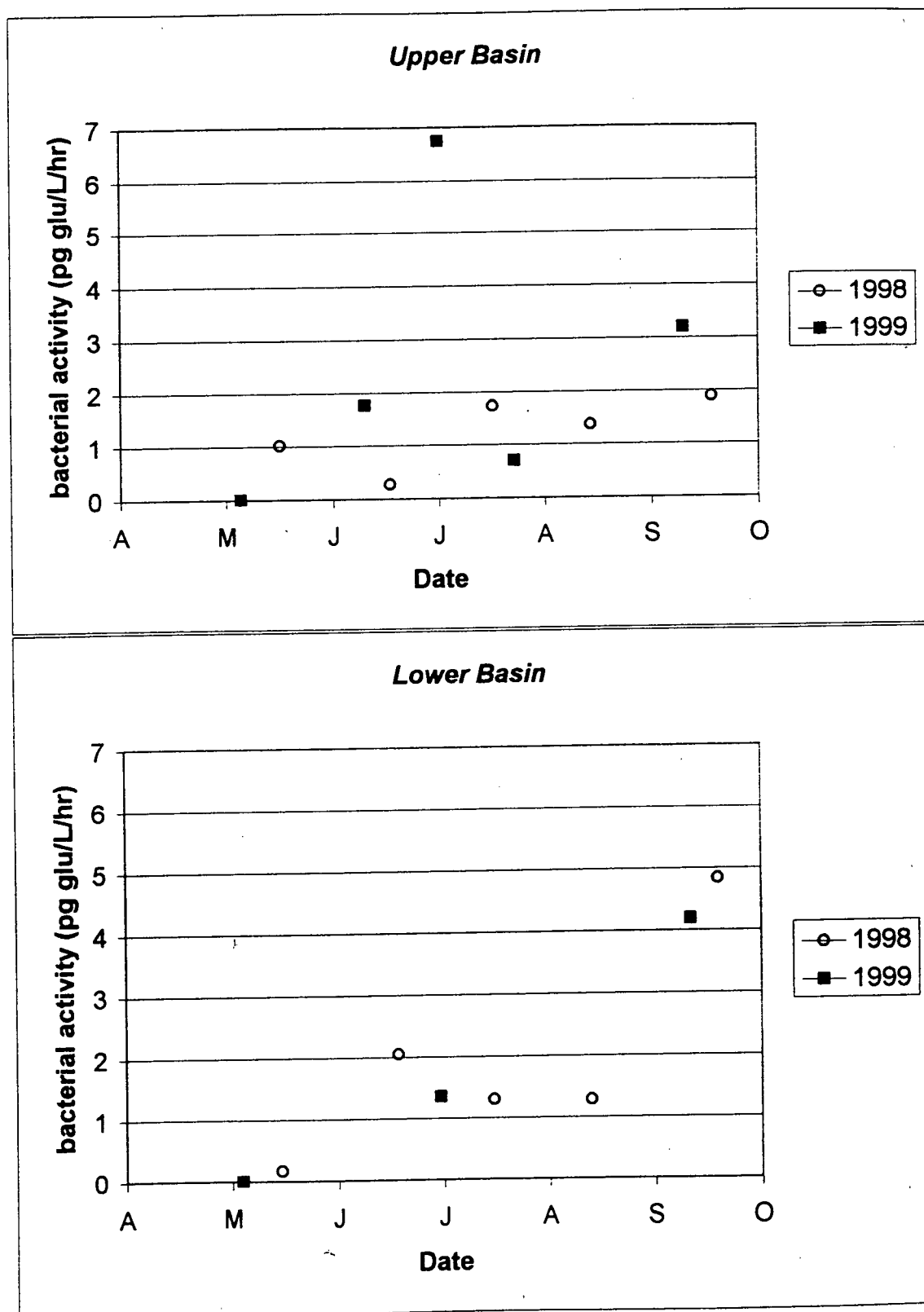


Figure 4.9 - Arrow Reservoir monthly bacterial abundance profiles

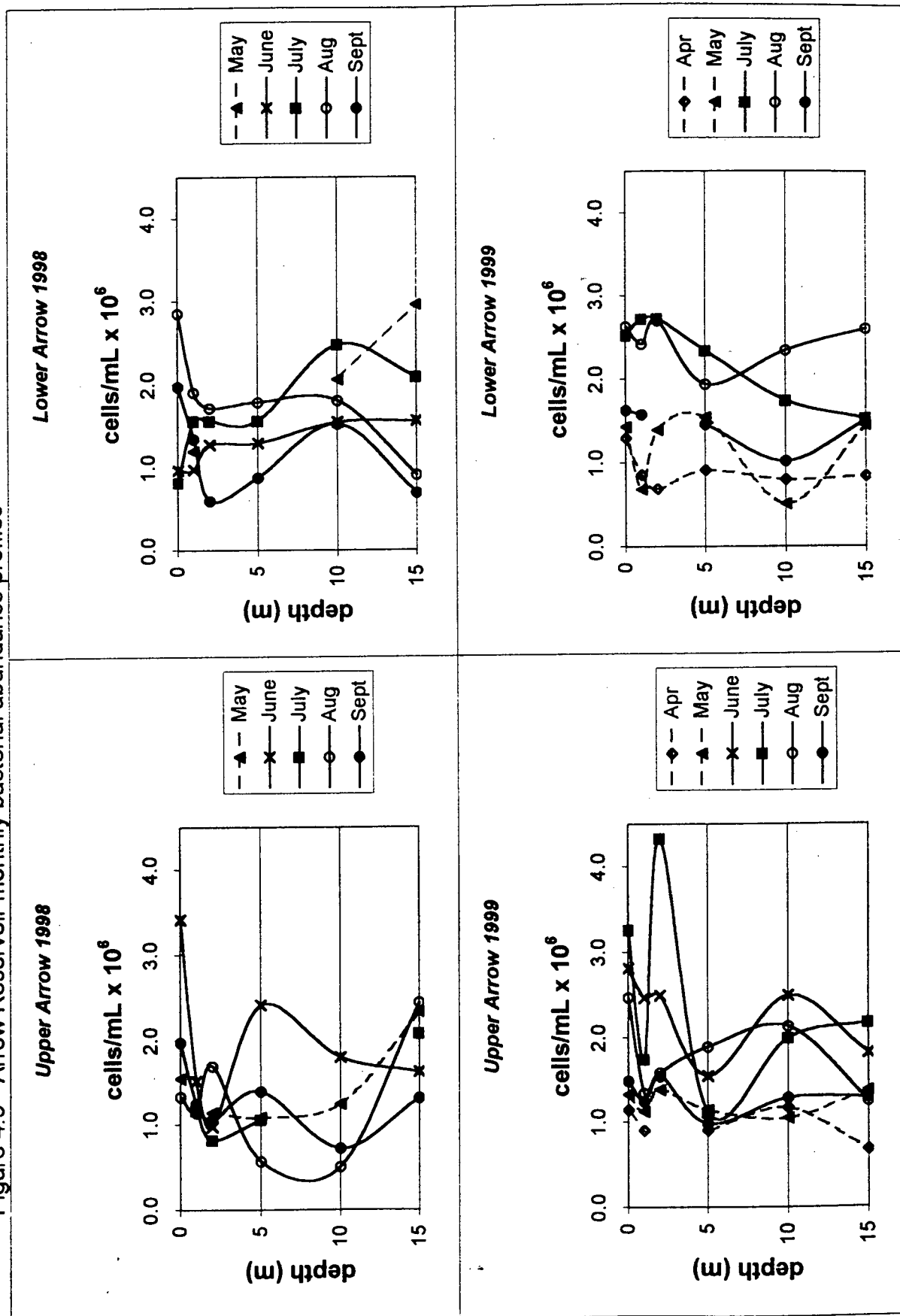




Table 4.5 Summary of the Arrow Reservoir bacterial abundance results

| Year | Basin | Depth weighted average bacterial abundance (cells/mL x 10 <sup>6</sup> ) |      |      |      |      |       |
|------|-------|--|------|------|------|------|-------|
|      |       | April  | May  | June | July | Aug. | Sept. |
| 1999 | Upper | 0.95   | 1.23 | 2.27 | 2.43 | 1.78 | 1.30  |
|      | Lower | 0.90   | 1.16 | -    | 2.26 | 2.44 | 1.44  |
| 1998 | Upper | -  | 1.47 | 1.95 | 1.28 | 1.28 | 1.28  |
|      | lower | -  | 2.06 | 1.26 | 1.67 | 1.82 | 1.16  |

"-" No samples collected during this period

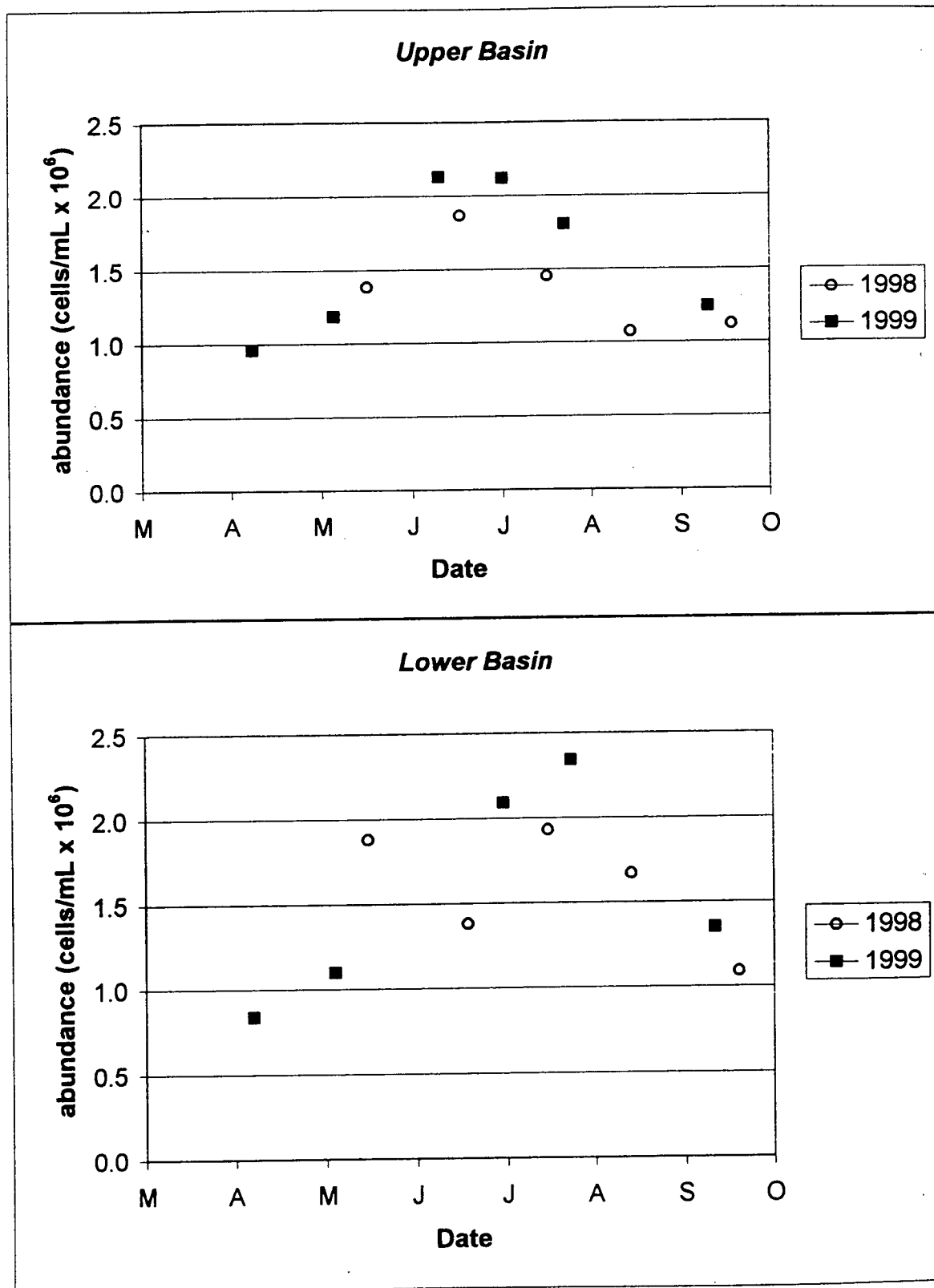
There was no statistical difference in cell abundance in the two basins in 1999 (p value = 0.4), nor was there any difference between the two years in the lower basin (p value = 0.4). The cell abundance in the upper basin in 1999 was significantly greater than that in the same basin in 1998 (p value = 0.04). In 1998 the difference between the lower basin and the upper basin was significant at a p value of 0.05. In this case, the lower basin was slightly greater than the upper basin.

The weighted average cell abundance was calculated for each month based on the relative size of the intervals between the sampling depths. An outline of the calculations used for this method is provided in Appendix 1. The weighted average bacterial abundance seasonal profile is presented in Figure 4.10. From a visual inspection of this figure, it appears that the difference in the abundance of bacteria between basins in a given year is relatively small. However, from a comparison of the two years, it appears that the cell abundance was lower in 1999 near the start of the season and higher in 1999 during the peak summer period and near the end of the season in September.

#### 4.1.5 CTC

As discussed in the methodology section, this method was implemented in July 1998 as an alternative to the unsuccessful INT method. Data is only available for the month of August in 1998. In 1999, this experiment was not performed in the lower basin in June. In addition, due to an insufficient incubation period in July of 1999, data for this parameter is not available for this month. The monthly depth weighted average of the

Figure 4.10 Seasonal bacterial abundance profiles for the Arrow Reservoir



percentage of active cells in the lower and upper basins of the reservoir was calculated based on the relative size of the intervals between the sampling depths. This data is presented in Table 4.6.

Table 4.6 - Summary of Arrow Reservoir CTC results

| Year                          |             | 1998 | 1999 |     |      |      |      |       |
|-------------------------------|-------------|------|------|-----|------|------|------|-------|
| Month                         |             | Aug. | Apr. | May | Jun. | Jul. | Aug. | Sept. |
| Percentage of active bacteria | Upper basin | 3.8  | 8.0  | 4.1 | 5.0  | -    | 2.4  | 1.6   |
|                               | Lower basin | 4.3  | 7.6  | 4.4 | -    | -    | 0.68 | 1.1   |

"-" No data available for this period

The percentage of active bacteria was greater in the spring than in late summer in both basins in 1999. In addition, the percentage of active bacteria is generally slightly higher in the upper basin than the lower basin in 1999. In August of 1998, the percentage of active bacteria was greater in the lower basin than the upper basin.

#### 4.1.6 Alkalinity

The alkalinity was measured primarily for use in the primary productivity calculations. In general, this parameter did not change with depth. However, on a few occasions, the alkalinity varied with depth and a separate equation was used to calculate the primary productivity at different depths. The monthly alkalinity in each basin in 1998 and 1999 is presented in Table 4.7. Where the alkalinity varies with depth, an average value, marked with an asterisk, is presented in this table.

Table 4.7 - Monthly alkalinity in the upper and lower basins of the Arrow Reservoir

| Year | Basin       | Monthly alkalinity (mg CaCO <sub>3</sub> /L) |      |       |      |        |       |
|------|-------------|--|------|-------|------|--------|-------|
|      |             | April  | May  | June  | July | August | Sept. |
| 1998 | Upper basin | -  | 49.0 | 45.6* | 43.7 | 39.6*  | 47.4* |
|      | Lower basin | -  | 47.6 | 46.2  | 44.6 | 43.5   | 41.5* |
| 1999 | Upper basin | 50.0   | 54.0 | 48.0  | 47.8 | 43.0   | 38.7  |
|      | Lower basin | 53.6   | 53.0 | -     | 44.9 | -      | 36.9  |

"-" no samples were collected during this period

In general, the alkalinity was greater in the spring than the summer and early fall. There is no apparent difference between the alkalinity in the upper and lower basins.

## 4.2 Horizontal Transects

Horizontal transects were performed in June, August and September 1999 in the upper basin of the Arrow Reservoir. The location of the transect stations is shown in Figure 3.1. As outlined in Section 3.2, stations 1 through 6 were chosen such that they are positioned in a north-south transect of the basin approximately equidistant from one another.

### 4.2.1 Primary Productivity

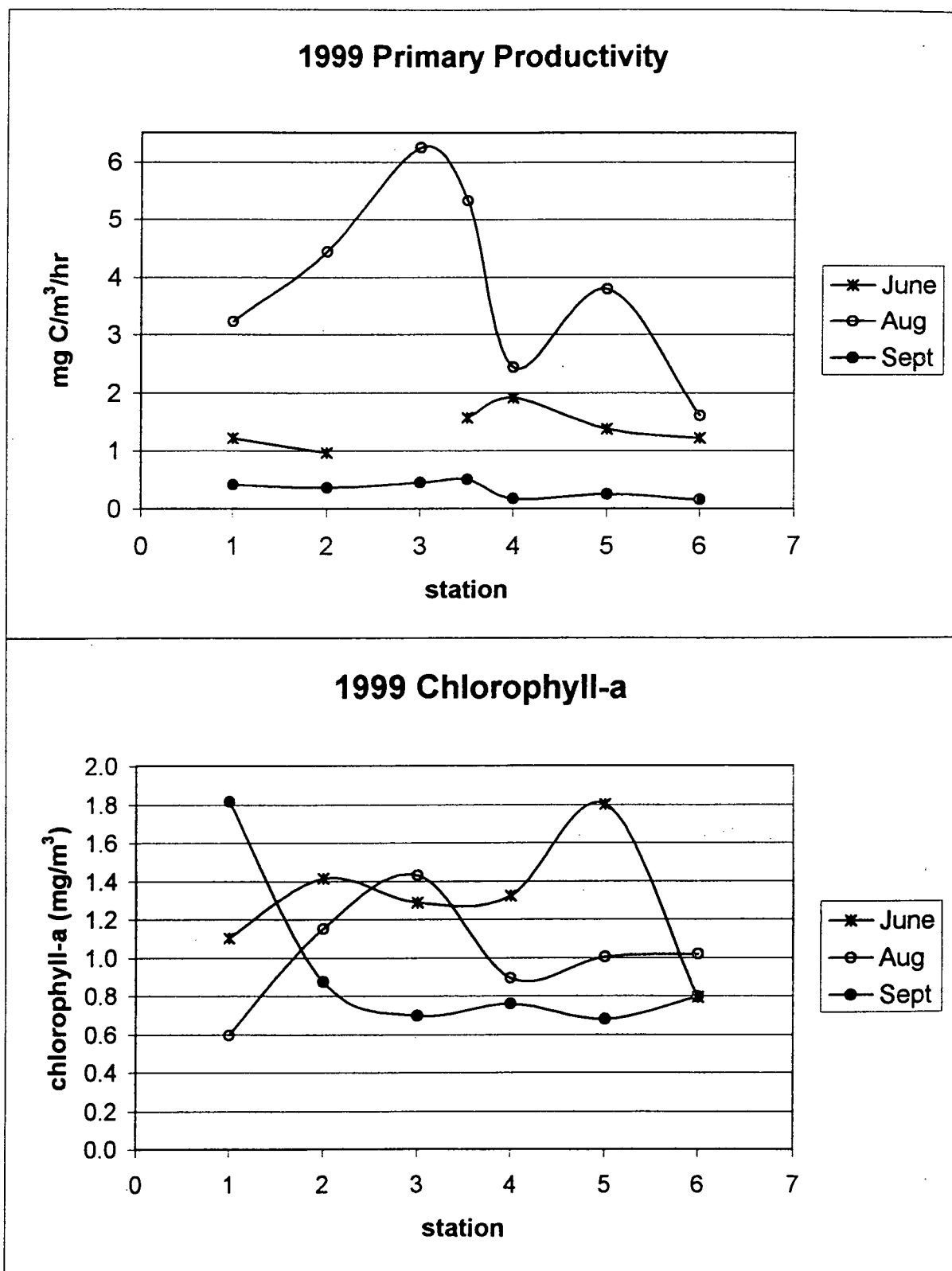
In June, the primary productivity peaked at station 4 ( $1.91 \text{ mg C/m}^3/\text{hr}$ ), approximately mid-way down the upper basin. The primary productivity at the southern end of the basin ( $1.22 \text{ mg C/m}^3/\text{hr}$ ), near Nakusp, was equivalent to that at the northern end of the basin, near Galena Bay.

Two primary productivity peaks were observed in the transect profile in August. The greatest primary productivity was observed at station 3 ( $6.26 \text{ mg C/m}^3/\text{hr}$ ), while a second smaller peak was recorded at station 5 ( $3.79 \text{ mg C/m}^3/\text{hr}$ ). The lowest productivity ( $1.60 \text{ mg C/m}^3/\text{hr}$ ) was recorded at station 6, near the southern end of the basin. The primary productivity was significantly higher in this month than in June (students t-test,  $p$  value = 0.003, 95% confidence level) and September (students t-test,  $p$  value < 0.001, 95 % confidence level).

In September, the transect primary productivity was significantly lower than in June (students t-test,  $p$  value < 0.001, 95 % confidence level) and in August. As in August, the maximum productivity was recorded at station 3 ( $0.50 \text{ mg C/m}^3/\text{hr}$ ), while a second smaller peak was observed at station 5 ( $0.25 \text{ mg C/m}^3/\text{hr}$ ) and the lowest productivity ( $0.15 \text{ mg C/m}^3/\text{hr}$ ) was measured at station 6.

The transect primary productivity profiles are depicted in Figure 4.11.

Figure 4.11 Horizontal transect primary productivity and chlorophyll-a profiles



#### 4.2.2 Chlorophyll-a

In June, chlorophyll-a varied from 0.79 mg/m<sup>3</sup> at station 6 to 1.80 mg/m<sup>3</sup> at station 5. The range of chlorophyll-a in the August transect was 0.60 mg/m<sup>3</sup> at station 1 to 1.43 mg/m<sup>3</sup> at station 3. In September, the maximum value of chlorophyll-a was 1.82 mg/m<sup>3</sup> at station 1. The range of values for the remaining stations was 0.68 mg/m<sup>3</sup> (station 5) to 0.88 mg/m<sup>3</sup> (station 2). The transect chlorophyll-a profiles are presented in Figure 4.12. This figure reveals that the overall variability of chlorophyll-a from month to month was relatively small. However it appears that the peak value gradually shifted from the south of the basin in June to the north end in September.

#### 4.3 Containment Experiments

Containment experiments were performed in April, May and July 1999. The addition of nutrients to carboy 2 was designed such that the nutrient concentrations and N:P ratio in the carboy would be approximately equivalent to that in the lake at that time as a result of fertilization. Carboy 1 received a lower N:P ratio, while carboy 3 received a higher ratio. The concentration of phosphorus was the same in all three carboys. The control carboy did not receive any nutrients. The exact nutrient concentrations and N:P ratios in each carboy for each of the three experiments are given in Table 3.1.

The purpose of the containment experiments was to determine whether the N:P ratio of the fertilizer at a given time was optimal for primary production. The results of the first two containment experiments in April and May were somewhat questionable. It is likely that zooplankton grazing of the phytoplankton affected the results of these experiments. It is also possible that some compound, toxic to phytoplankton, was present in the sample water, which was collected from a marina with fairly regular boat traffic.

Several changes were made for the third and final containment experiment, which was performed in July. In order to avoid contamination with toxic substances, sample water was collected from a region of the marina where boat traffic was at a minimum. In addition, sample water was filtered through a plankton net prior to filling the carboys to eliminate potential differential grazing pressures within the carboys. The final change in

the experiment in July involved the volume of nutrient solution added to each carboy. In the first two experiments, the volume of nutrient solution added was 1 mL. In July, the appropriate masses of nitrogen and phosphorus were dissolved into 100 mL and this significantly larger volume was added to the carboys. The decision to add the nutrients in a larger volume of water was based on the expectation that this would perhaps allow for better dispersion of the nutrients within the carboys.

The results of the experiment in July were promising. The primary productivity in carboy 2 (N:P 6.00), the carboy with the same N:P ratio as would be expected in the lake at that time due to fertilizer addition, was significantly greater than that in each of the other carboys. In addition, the greatest chlorophyll-a of the four carboys was measured in carboy 2. The chlorophyll-a in this carboy was slightly greater than that in carboy 3 (N:P 7.50) and more than double that in carboy 1 (N:P 4.50) and the control.

#### 4.3.1 Primary Productivity

The results of the primary productivity analyses are presented in Table 4.8. In May, no valid results were obtained. The radioactive counts in the dark bottle were greater than those in one of the light bottles in carboy 1, two of the light bottles in carboy 2 and all three of the light bottles in carboy 3 and the control carboy.

Table 4.8 - Containment experiment primary productivity results

| month | Carboy productivity (ug C/L/hr) |      |      |         |
|-------|---------------------------------|------|------|---------|
|       | 1                               | 2    | 3    | Control |
| April | 1.64                            | 1.10 | 1.38 | 0.78    |
| July  | 1.45                            | 3.31 | 1.23 | 1.21    |

There was no statistical difference in the mean productivity in the four carboys in April. In July, the productivity in carboy 2 was significantly greater than that in carboys 1 (N:P 4.5), 3 (N:P 7.5) and the control.

#### 4.3.2 Chlorophyll-a

The results of the chlorophyll-a analyses are presented in Table 4.9.

Table 4.9 - Containment experiment chlorophyll-a results

| Month | Carboy chlorophyll-a (mg/m <sup>3</sup> ) |      |      |         |
|-------|---|------|------|---------|
|       | 1   | 2    | 3    | control |
| April | 0.30                                      | 0.47 | 0.30 | 0.49    |
| May   | 0.38                                      | 0.12 | 0.03 | 0.17    |
| July  | 0.69                                      | 1.42 | 1.27 | 0.47    |

No statistical analyses could be performed with this data as there was no replication. In April, the chlorophyll-a in carboys 1 (N:P 0.45), 2 (N:P 0.67) and 3 (N:P 1.50) was lower than that in the control carboy, while in May, only the chlorophyll-a in carboy 1 was greater than that in the control. In July, the chlorophyll-a in the control carboy was lower than that in each of the other carboys. The chlorophyll-a in carboy 2 (N:P 6) was the highest and it was more than double that in carboy 1 (N:P 4.5) and the control. The chlorophyll-a in carboy 3 (N:P 7.5) was slightly lower than that in carboy 2.

#### 4.4 Stream Bioassays

Bioassays were performed on samples collected from tributaries during spring freshet and summer low flows. These samples were collected by Grant Thorp, Diana Koller and Dean den Biessen and samples were simultaneously collected for nutrient analyses. The results of the nutrient analyses are summarized in Table 4.10, where NH<sub>3</sub> is ammonia, NN is nitrate nitrogen, OP is orthophosphate, TDP is total dissolved phosphorus, TP is total P and N:P is the nitrogen to phosphorus ratio. Full bioassays were performed on eight samples and two modified bioassays were performed on a total of 16 samples.



Table 4.10 – Summary of stream water quality

| Location | Date     | NH <sub>3</sub> ** | NN** | OP*** | TDP*** | TP*** | N:P <sup>x</sup> |
|----------|----------|--------------------|------|-------|--------|-------|------------------|
| Inono.   | 05/19/98 | 8                  | 17   | 1     | 5      | 41    | 5                |
| Inono.   | 08/10/98 | 5                  | 3    | 1     | 7      | 12    | 1                |
| Incom.*  | 05/26/98 | 5                  | 353  | 1     | 2      | 12    | 179              |
| Incom.   | 09/08/98 | 5                  | 57   | 1     | 2      | 59    | 31               |
| Mosqu.   | 05/19/98 | 15                 | 89   | 1     | 4      | 59    | 26               |
| Mosqu.   | 09/08/98 | 5                  | 4    | 1     | 4      | 12    | 2                |
| Burton   | 06/01/98 | 7                  | 38   | 1     | 2      | 7     | 22               |
| Half.    | 06/01/98 | 5                  | 102  | 1     | 3      | 3     | 36               |
| What.    | 05/19/98 | 5                  | 12   | 1     | 3      | 4     | 6                |
| What.    | 08/10/98 | 5                  | 57   | 1     | 2      | 5     | 31               |
| Kusk.    | 06/01/98 | 5                  | 42   | 1     | 6      | 14    | 8                |
| Kusk.    | 08/10/98 | 5                  | 48   | 1     | 6      | 6     | 9                |
| Carib.   | 05/19/98 | 5                  | 138  | 1     | 3      | 9     | 48               |
| Carib.   | 09/08/98 | 23                 | 113  | 2     | 7      | 18    | 19               |
| Half.    | 08/10/98 | 5                  | 32   | 1     | 11     | 6     | 3                |
| Burton   | 09/08/98 | 15                 | 81   | 1     | 3      | 21    | 32               |
| Eagle    | 05/19/98 | 5                  | 19   | 1     | 4      | 6     | 6                |
| Eagle    | 08/10/98 | 7                  | 22   | 1     | 7      | 12    | 4                |
| St Leon* | 06/01/98 | 5                  | 73   | 1     | 3      | 4     | 26               |
| St Leon  | 08/10/98 | 5                  | 70   | 1     | 6      | 6     | 13               |
| Syringa* | 06/05/98 | 8                  | 8    | 3     | 4      | 9     | 4                |
| Syringa  | 08/05/98 | 5                  | 3    | 1     | 17     | 19    | 0.5              |
| Deer*    | 06/05/98 | 5                  | 2    | 10    | 13     | 5     | 0.5              |
| Colum.*  | 06/06/98 | 5                  | 105  | 4     | 2      | 4     | 55               |

<sup>x</sup> ratio of total dissolved inorganic nitrogen (NH<sub>3</sub> + NN) to total dissolved phosphorus (TDP)

\*\* water quality sample collected on 05/19/98, except Colum. which was collected 06/01/98

\*\*\* units are ug/L as N

\*\*\*\* units are ug/L as P

Note – Full bioassays were performed on the first 8 samples; modified bioassays were performed on the remaining samples.

#### 4.1.1 Full bioassays

Full bioassays were performed on the following 8 samples: Inonoaklin Creek (May), Inonoaklin Creek (August), Mosquito Creek (May), Mosquito Creek (September), Incomappleux (May), Incomappleux (September), Halfway River (June), Burton Creek (June).

Algal growth was assessed with daily coulter counter and fluorometer readings. However, there was a significant discrepancy between the results obtained by these two methods. With each sample, fluorometer readings indicated a substantial net growth only

in tubes spiked with a combination of N and P. In contrast, the coulter counter readings indicated a substantial net growth in tubes spiked with a combination of N and P or with P but not N. Since the coulter counter measures the number of particles in a mL of sample it is possible that there is some other "particle" formation in the samples spiked with P. Several tests were performed to determine the nature of these additional particles.

Samples spiked with N only and P only were stained with DAPI and examined microscopically to determine whether there was a significantly greater quantity of bacteria present in the samples spiked with P. The abundance of bacteria present in the samples spiked with P was not greater than that in samples spiked with N and was not deemed great enough to generate the larger particle numbers observed with the coulter counter.

Samples spiked with N only and P only were also examined microscopically to determine whether a precipitate had formed in the samples spiked with P. No particles in addition to the *S. capricornutum* cells were observed.

A bioassay was performed in which additional controls were used: sample spiked with N + P (no *S. capricornutum* was added); distilled deionized water spiked with *S. capricornutum* and N+P+micronutrients. No significant growth was observed in these controls. Therefore, the additional particles in samples spiked with P were not likely a precipitate as this precipitate should have formed in these additional controls. Finally, the additional particles are not likely due to a contaminant organism in the P nutrient spikes as they should have grown in these additional controls.

Since the coulter counter measures particle abundance, while the fluorometer measured chlorophyll-a, the fluorometer was considered to be the more reliable method of measuring planktonic growth. Therefore, only the results of the fluorometer readings are presented and analysed in this thesis. Net growth was estimated based on the difference between the final and initial fluorometer readings. The results of the statistical analyses of the bioassay results are summarized in Table 4.11.

Table 4.11 – Summary of full bioassay statistical analyses

| sample        | f vs. nf*            | N+P vs. others*      | P values from statistical contrasts |                      |                      |                      | Nu vs. w/o Nu*     |
|---------------|----------------------|----------------------|-------------------------------------|----------------------|----------------------|----------------------|--------------------|
|               |                      |                      | Ctrl vs. others*                    | N vs. Ctrl & P & Nu* | P vs. Ctrl & N & Nu* | N vs. P*             |                    |
| Incom (May)   | -                    | $9 \times 10^{-10}$  | -                                   | -                    | -                    | -                    | -                  |
| Incom (Sept.) | NA                   | $2.9 \times 10^{-6}$ | -                                   | -                    | -                    | -                    | -                  |
| Inono (May)   | -                    | $5 \times 10^{-10}$  | -                                   | $9 \times 10^{-7}$   | $1 \times 10^{-4}$   | $3.8 \times 10^{-7}$ | -                  |
| Inono (Aug.)  | -                    | $2 \times 10^{-26}$  | $1.8 \times 10^{-2}$                | $1.1 \times 10^{-5}$ | $8.4 \times 10^{-8}$ | $6.1 \times 10^{-8}$ | -                  |
| Mosqu (May)   | -                    | $3 \times 10^{-16}$  | -                                   | -                    | -                    | -                    | -                  |
| Mosqu (Sept.) | $9.9 \times 10^{-3}$ | $7 \times 10^{-28}$  | -                                   | $3 \times 10^{-14}$  | $4 \times 10^{-12}$  | $6 \times 10^{-15}$  | $9 \times 10^{-4}$ |
| Half (June)   | -                    | $9.8 \times 10^{-8}$ | -                                   | -                    | -                    | -                    | -                  |
| Burton (June) | -                    | $7 \times 10^{-15}$  | -                                   | -                    | -                    | -                    | -                  |

"-" no statistical significance - P value greater than 0.05

NA – not applicable: no results available for filtered samples

"\*" see methods (statistical analyses) section for explanation

The results of these analyses were similar for samples collected from Incomappleux River in May and September, Halfway River in June and Burton Creek in June. In each of these samples, only tubes to which N and P were added in combination exhibited a significant increase in growth. There was no significant difference in growth between filtered and unfiltered sample. The addition of micronutrients to the samples made no significant difference in algal growth.

Samples from Innonoaklin in May and August and Mosquito Creek in September also experienced a much greater net growth with the addition of N and P than with all other treatments. In addition, a slightly enhanced net growth was observed in these samples with the addition of N only, indicating a slight N limitation.

Only the sample collected from Mosquito Creek in September was influenced by filtration or the addition of micronutrients.

#### 4.1.2 Modified bioassays

Two separate modified bioassays were performed in which 8 different stream samples were assayed at once. These two modified bioassays will be treated separately as two separate *Selenastrum* cultures were utilized for sample inoculation. In addition, the initial abundance of *Selenastrum* was slightly different in the two bioassays.

The following samples were assayed in the first bioassay: Whatshan Creek (May 1998), Whatshan Creek (August 1998), Kuskanax River (June 1998), Kuskanax River (August 1998), Caribou Creek (May 1998), Caribou Creek (September 1998), Halfway River (August 1998), Burton Creek (September 1998). The second bioassay was performed on the following samples: Eagle Creek (May 1998), Eagle Creek (August 1998), St. Leon River (June 1998), St Leon (August 1998), Syringa Creek (June 1998), Syringa Creek (August 1998), Deer Creek (June 1998) and Columbia River (June 1998).

As would be expected, for each sample, the addition of N and P resulted in a significantly larger net growth. However, the net growth in samples to which N and P were added was highly variable. The results of the two modified bioassays are presented in Tables 4.12 and 4.12 along with the N:P ratio for each sample.

Table 4.12 - Summary of the results of the first modified bioassay

| Net growth | What. May | What. Aug | Kusk. June | Kusk. Aug | Carib. May | Carib. Sept | Half Aug | Burton Sept |
|------------|-----------|-----------|------------|-----------|------------|-------------|----------|-------------|
| Sample*    | 17.2      | 13.5      | 14.8       | 17.8      | 18.3       | 17.6        | 17.8     | 9.9         |
| N+P        | 61.2      | 119.5     | 83.4       | 43.7      | 71.5       | 101.4       | 115.8    | 130.7       |
| N:P**      | 6         | 31        | 8          | 9         | 48         | 19          | 3        | 32          |

Results are in fluorescence units

\* "sample" indicates raw sample without added nutrients

\*\* ratio of total nitrogen (NN + NH<sub>3</sub>) to total dissolved phosphorus (TDP)

Table 4.13 - Summary of the results of the second modified bioassay

| Net growth | Eagle May | Eagle Aug | St.Leon June | St.Leon Aug | Syringa June | Syringa Aug | Deer June | Colum. June |
|------------|-----------|-----------|--------------|-------------|--------------|-------------|-----------|-------------|
| Sample*    | 9.8       | 25.4      | 17.6         | 23.2        | 15.9         | 18.4        | 28.6      | 30.4        |
| N+P        | 59.1      | 109.5     | 79.8         | 76.6        | 34.4         | 75.4        | 133.8     | 86.3        |
| N:P**      | 6         | 4         | 26           | 13          | 4            | 0.5         | 0.5       | 55          |

Results are in fluorescence units

\* "sample" indicates raw sample without added nutrients

\*\* ratio of total nitrogen (NN + NH<sub>3</sub>) to total dissolved phosphorus (TDP)

For each of the two modified bioassays, the relationship between the net growth in sample alone and in sample spiked with N and P was assessed with a regression analysis. There was a slight negative correlation between the net growth in sample alone and in enriched sample in the first bioassay with a slope of -6.8 and an  $r^2$  of 0.4. A slight positive correlation exists between the net growth in sample alone and in enriched sample in the second bioassay with a slope of 3.1 and an  $r^2$  of 0.5.

Regression analyses were also performed with the results of each of the two modified bioassays to assess the relationship between the net growth and the N:P ratio in the samples. There was no significant correlation between the net growth and the N:P ratios in the samples in the first and second modified bioassays with  $r^2$  values of 0.08 and 0.2, respectively.

## 5. DISCUSSION

### 5.1 Vertical transects

#### 5.1.1 Primary productivity

To better comprehend the nature of primary productivity profiles measured in the Arrow Reservoir during this study, it is important to understand the factors that affect phytoplankton and their productivity. Phytoplankton growth is dependent upon several variables, including light intensity, nutrient availability, water temperature and biotic factors, such as competition, predation and parasitism within the euphotic zone (Wetzel 1982). Available light for photosynthesis and its intensity is seasonally dependent both upon the time of year and time of day. The extent of light penetration into the surface water of the reservoir is a function of the turbidity of the water, the weather and water turbulence. Nutrients available for phytoplankton depend on the nutrient supply, i.e. incoming nutrients. Nutrients in particulate form, which have sunk below the euphotic zone, and are no longer available for photosynthesis, will either be sedimented or returned to the surface layers by deep mixing in early spring and late autumn.

Since light intensity decreases exponentially with depth, in the absence of other effects, the primary productivity would be expected to decrease exponentially with depth. However, in most relatively clear lakes, productivity is low near the surface and peaks at 3 – 5 m due to the deleterious effects of UV light on phytoplankton, a phenomenon referred to as photoinhibition (Horne and Goldman, 1994). This type of curve was commonly seen in Arrow Reservoir, particularly during times of peak insolation (May, June, July)

While the monthly productivity profiles in each basin and during each year follow a similar trend, there was considerable variation, due in part to photoinhibition but also to a variable compensation depth. In general, when the compensation depth is within 1 to 2 m below 15 m, the productivity at 15 m is less than at the surface. However, when the compensation depth is greater than 2 m below 15 m, the productivity at 15 m is usually greater than at the surface. In addition, deeper peak productivity is often observed with

deeper compensation depths. This provides evidence to indicate the presence of subsurface plates, i.e. deep chlorophyll maxima.

A comparison of the local flow data for the reservoir in 1998 with a historical average (1986-90, 95-97) is shown in Figure B.1. This plot reveals that the freshet began early in 1998, around the beginning of May. From early June until September, local flow was much lower than the historical average. Figures B 2, B 3, B 4 and B 5 in Appendix B reveal that the light penetration was not as deep during peak spring freshet in May and June when the input of suspended material was likely greatest. Following freshet, there was an increase in light penetration in both basins. In the lower basin where this increase was most dramatic, there was a steady decrease in phytoplankton biomass.

In 1999, a cooler cloudier spring delayed the start of freshet until the end of May. This can be seen in Figure B.1 where local flow in 1999 is compared with the historical average. A relatively large peak in local flow was observed in mid-June 1999 and the flow remained greater than the historical average until September. The compensation depths in April and May were relatively deep in both basins. During mid-summer, the compensation depths were unusually shallow in the upper basin, ranging from only 7.5 m in June to 11 m in August. These shallow compensation depths are likely due to high turbidity from a late freshet derived from a relatively large snow pack. In summary, the compensation depths in the lower basin were greater than those in the upper basin. With the exception of the unusually shallow compensation depths in the upper basin in mid-summer in 1999, the compensation depths in the two basins of Arrow Reservoir in 1998 and 1999 were typically at approximately 15 m or deeper.

Phytoplankton populations tend to follow a seasonal succession, which, in more productive mesotrophic or eutrophic lakes, is often characterized by two to three distinct blooms (Wetzel 1982, Horne and Goldman 1994). The first bloom occurs in the spring following spring mixing and the commencement of thermal stratification when plankton are no longer subjected to deep mixing and are exposed to sufficient sunlight. The spring bloom is usually dominated by diatoms, which are a cold tolerant species that possess a cell wall composed of silica. This bloom generally declines when silica becomes depleted and predation by zooplankton increases. Following a period of low productivity and/or biomass, populations of species which are more tolerant to zooplankton grazing

may build up to form a summer bloom. A third bloom may occur in the fall when nutrients become more readily available due to the onset of deep mixing.

In the Arrow Reservoir, there were only small shifts in phytoplankton species composition throughout the period of May to October during 1997, 1998 and 1999 (Pieters et al. 1998, Stockner unpublished data). In addition, there were no apparent blooms in the reservoir during either of these years (Figure B 6) and changes in phytoplankton biomass throughout the growing season were minimal. Pieters et al. (1998, unpublished data) report that the phytoplankton community of the Arrow Reservoir from 1997 to 1999 was composed primarily of species characteristic of ultra-oligotrophic lakes: the dominant species were the picoplankter *Synechococcus* and Chryso- and Cryptophyceae nano-flagellates. While diatoms were present, they did not show large increases in spring or fall. Profiles of the seasonal abundance and biomass of the major phytoplankton classes in the reservoir at 8 stations (stations 2 and 7 correspond to the upper and lower basin sampling locations for this thesis) from May 1998 to November 1999 are shown in Figures B 7 and B 8.

In 1998, daily areal primary productivity increased from May to a single peak in July and then decreased to a relatively low value in September of similar magnitude to that in May. In 1999, primary productivity measurements began in April, one month earlier than in 1998. Two daily areal primary productivity peaks were observed in each basin during this sampling season. In the lower basin, peaks of equal magnitude were observed in April and July. The first peak in the upper basin occurred in May while a second larger peak was observed from July to August.

Since the weather conditions on a given sampling day may not be representative of those for the period during which the samples were collected, the "monthly" averaged areal primary productivity (expressed as a daily rate) may be a useful parameter for monthly comparisons. However, one should bear in mind that significant changes in primary productivity in response to changes in nutrient availability or biotic factors may occur over a short period of time. In 1998, the daily areal primary productivity and the corresponding monthly average areal primary productivity were very similar. Therefore, the weather conditions on the various sampling dates were fairly representative of those for the period surrounding the sampling dates. In 1999, the trends in daily areal primary



productivity and monthly average areal primary productivity were similar. However, while the peak in daily areal primary productivity observed in the summer was greater than that observed in the spring, the summer peak in monthly average areal primary productivity was smaller than the spring peak. The daily areal primary productivity and monthly average areal primary productivity in the reservoir are depicted in Figures 4.3a and 4.3b, respectively.

The primary productivity peak observed in the spring of 1999 was likely in response to increasing total solar radiation. During April 1999 a broad, relatively deep peak (from approximately 5 to 15 m) in primary productivity was observed in both basins. No significant thermal stratification had begun when these samples were collected (Figure B 9).

The decline in primary productivity in late spring/early summer may be attributable in part to decreasing nutrient concentrations as nutrients are bound up in phytoplankton biomass. The low light intensity and the unusually shallow compensation depth of 7.5 m in the upper basin in June may also have played a role in the low primary productivity observed during this month. Since lake fertilization began immediately following the April primary productivity measurements, the extended productivity peak observed in the upper basin is likely a function of the additional nutrients. Had the primary productivity of the reservoir been measured in April 1998, it is likely that a spring peak would have been observed in each basin.

Since there was little change in the phytoplankton species composition in the reservoir, the increase in primary productivity in the early summer is not due to a succession of species. Nutrient inputs from the freshet, increasing sunlight intensity and increasing light penetration as freshet declined likely all play a role in the increase in primary productivity observed in early to mid summer.

In late summer, the decrease in primary productivity is likely due in part to the decreasing sunlight intensity and daylight hours. During early September the daily solar radiation declined rapidly in both 1998 and 1999 (see Figure B 10). Predation, parasitism and nutrient limitation may also be factors in the decline in primary productivity.

The primary productivity of a sample is a function of the number of cells in the sample and the productivity of those cells. Thus, a smaller number of more productive

cells may have the same primary productivity as a larger number of less productive cells. While primary productivity is a measure of the total productivity of the autotrophic phytoplankton community, the specific growth rate (primary productivity per unit chlorophyll-a) provides an indication of the efficiency of carbon production, i.e. photosynthesis.

The seasonal trends in specific growth rate in Arrow Reservoir during the period of May to September 1998 and the period of April to September in 1999 are depicted in Figure 4.4. The specific growth rates in the two basins were very similar in 1998 and they were smaller than those observed in 1999. The highest specific growth rates during this study were observed in the lower basin in April and July of 1999.

The specific growth rates in the Arrow Reservoir in 1998 ranged from 0.20 to 2.03 mg C/mg chlorophyll-a/hour in the lower basin and 0.36 to 2.08 mg C/mg chlorophyll-a/hour in the upper basin. In 1999, the specific growth rates in the lower and upper basins varied from 0.35 to 4.25 and 0.27 to 3.23 mg C/mg chlorophyll-a/hour, respectively. Stockner (1987) reports the following average specific growth rates (reported as P/B ratios with the units mg C/mg chlorophyll-a/day) in BC lakes: 6.3 in unfertilized north coastal, 6.8 in unfertilized south coastal, 14.2 in fertilized north coastal, 7.8 in fertilized south coastal and 10.7 in interior lake(s). In this context the specific growth rates in the Arrow Reservoir are relatively low.

When factors such as climate and nutrient availability vary from one year to the next, primary productivity will also show annual variation. No perceptible differences in the solar radiation (the RMS between the lower and upper basins for the 15 day running mean was  $15.09 \text{ W/m}^2$  in 1998 and  $9.98 \text{ W/m}^2$  in 1999) nor in air temperature (the RMS between the lower and upper basins for the 15 day running mean was  $0.57^\circ\text{C}$  in 1998 and  $0.55^\circ\text{C}$  in 1999) were observed between the two basins of the Arrow Reservoir, which suggests that the weather patterns are very similar in the two basins. In the absence of human interference within the Arrow Reservoir, most natural annual variations in primary productivity would likely be experienced to some extent within both basins. Figures B 11 and B 10 reveal that the spring and summer of 1998 were relatively warm and sunny, while in 1999 they were relatively cool and overcast. If a change in climate from 1998 to 1999 influenced the productivity in the reservoir it would likely have

resulted in a lower productivity in each basin. The primary productivity in the upper basin increased during the first season of fertilization in this basin. Due to the lack of a parallel change in productivity in the lower basin (where the fertilizer may not yet have had any influence), it is likely that the variation in productivity in the upper basin is due to fertilizer addition.

A limnological study of four BC reservoirs, including Arrow Reservoir, was performed in September 1976 (Malick 1977). The primary productivity and chlorophyll-a profiles of the four reservoirs (Arrow, McNaughton, Upper Campbell and Williston) were estimated at a single location near the dam in each reservoir. The Arrow Reservoir primary productivity profile near the Keenleyside dam reached a peak of 1.5 mg C/m<sup>3</sup>/hour at a depth of 2 m. The profile in September 1976 was similar to those observed in the two basins of this reservoir in September 1998 and 1999. The peak values during these two years were less than half of that in 1976. In 1998, the maximum primary productivity in the upper basin was 0.68 mg C/m<sup>3</sup>/hour at 5 m and that in the lower basin was 0.67 mg C/m<sup>3</sup>/hour at 2 m. The peak primary productivity in 1999 was observed at 2 m (0.50 mg C/m<sup>3</sup>/hour) in the upper basin and from 1 and 5 m (0.54 to 0.57 mg C/m<sup>3</sup>/hour) in the lower basin.

The daily areal primary productivity and the maximum, minimum and mean chlorophyll-a in each of the four reservoirs in September 1976 are presented in Table 5.1 along with those of the Arrow Reservoir in September 1998 and 1999. While the daily areal primary productivity in the Arrow Reservoir in September 1998 and 1999 falls within the range observed in the four reservoirs in September 1976, it is much lower than that recorded in the Arrow Reservoir in 1976. The chlorophyll-a of these reservoirs in comparison with that of the Arrow Reservoir will be discussed in section 5.1.2.

Table 5.1 – Comparison of the September 1976 limnological study of four BC reservoirs with the September 1998 and 1999 data from the Arrow Reservoir

| Year | Reservoir      | Chlorophyll-a (mg/m <sup>3</sup> ) |              | Primary Productivity (mg C/m <sup>2</sup> /day) |
|------|----------------|------------------------------------|--------------|---|
|      |                | Mean                               | Range        |   |
| 1976 | Williston      | 2.96                               | 2.66 - 3.22  | 102   |
|      | Lower Arrow    | 0.92                               | 0.39 - 1.68  | 92.7  |
|      | Upper Campbell | 0.91                               | 0.44 - 2.55  | 10.7  |
|      | McNaughton     | 0.55                               | <0.10 - 0.76 | 59.4  |
| 1998 | Upper Arrow    | 0.96                               | 0.74 – 1.10  | 56.0  |
|      | Lower Arrow    | 1.05                               | 0.84 – 1.29  | 49.2  |
| 1999 | Upper Arrow    | 0.98                               | 0.30 – 1.16  | 27.8  |
|      | Lower Arrow    | 1.20                               | 0.88 - 1.65  | 56.5  |

Wetzel (1983) reports the mean daily productivity for an entire year for selected freshwater lakes of varying trophic status. This parameter varied from 1.6 to 249 mg C/m<sup>2</sup>/day in oligotrophic lakes, 210 to 729 mg C/m<sup>2</sup>/day in mesotrophic lakes and 820 to 1750 mg C/m<sup>2</sup>/day in eutrophic lakes. As a general rule, lakes with a mean primary productivity of less than 50 mg C/m<sup>2</sup>/day are ultraoligotrophic, those in the range of approximately 50 to 300 mg C/m<sup>2</sup>/day are considered to be oligotrophic, those in the range of approximately 250 to 1000 mg C/m<sup>2</sup>/day are mesotrophic and those with a mean primary productivity greater than 1000 mg C/m<sup>2</sup>/day are eutrophic (Wetzel 1983). The mean daily productivity for the growing season in the Arrow Reservoir was 133.2 mg C/m<sup>2</sup>/day in the upper basin and 146.0 mg C/m<sup>2</sup>/day in the lower basin in 1998 and 216.4 mg C/m<sup>2</sup>/day in the upper basin and 120.2 mg C/m<sup>2</sup>/day in the lower basin in 1999. These values, which fall in the oligotrophic range, are representative of the growing season only. Since the primary productivity is expected to be very low throughout the winter, the annual average would likely be substantially lower than that of the growing season and may fall within the ultra-oligotrophic range.

The primary productivity of many ultra-oligotrophic coastal lakes in BC was assessed as part of an extensive fertilization study. Stockner and Shortreed (1985) report average primary production during stratification, from April to October, ranging from 3.0 to 10.6 mg C/m<sup>3</sup>/day in unfertilized lakes and from 10.4 to 110.2 mg C/m<sup>3</sup>/day in fertilized lakes. Prior to fertilization, the average primary productivity in the Arrow reservoir (in mg C/m<sup>3</sup>/day) fell within or just above the range observed in the unfertilized

ultra-oligotrophic coastal lakes. During the first year of fertilization, the weighted average daily primary productivity in the Arrow Reservoir was generally close to the low end of the range measured in the fertilized coastal lakes. In 1998, prior to fertilization, the weighted average primary productivity varied from 3.7 to 16.7 mg C/m<sup>3</sup>/day in the upper basin and from 2.7 to 26.2 mg C/m<sup>3</sup>/day in the lower basin. During the first year of fertilization, the weighted average primary productivity ranged from 1.9 to 26.1 in the upper basin and 1.4 to 13.5 mg C/m<sup>3</sup>/day in the lower basin.

### 5.1.2 Chlorophyll-a

The trends in chlorophyll-a (phytoplankton biomass) are highly variable between years and basins in the Arrow Reservoir. In 1998, there was no significant difference ( $P = 0.1$ ) in the chlorophyll-a from the upper to the lower basin. In 1999, the chlorophyll-a in the lower basin was significantly lower ( $P = 0.02$ ) than in 1998. In contrast, no significant difference ( $P = 0.2$ ) was observed in the upper basin in 1998 and 1999.

In 1998, the chlorophyll-a in the reservoir was higher in the spring than in late summer/early fall. However, in 1999, the chlorophyll-a was lowest in the spring and highest in late summer/early fall in both basins. Some of this annual variation is likely due to climatic differences between years. Figures 5.5 and 5.6 show that, while the spring and summer of 1998 were relatively warm and sunny, in 1999 the spring and early summer were unusually cold and overcast.

There were no significant blooms in either basin in 1998 with the maximum abundance less than double the minimum abundance. Similar results were reported by Stockner (Pieters et al. 1998) for this reservoir in 1997. During the 1997 growing season, the average phytoplankton cell densities, biovolume and total carbon concentrations were greater in the lower basin than in the upper basin. However, in 1998 mean phytoplankton abundance and biomass was greater in the upper basin of the reservoir (Stockner, unpublished data). Based on the chlorophyll-a results, there was no significant difference ( $P = 0.1$ ) in the phytoplankton biomass in the upper and lower basins in 1998.

The variation between the minimum and maximum abundance was slightly larger in the reservoir in 1999 with a greater than 3 fold difference in the upper basin and a greater than 4 fold difference in the lower basin. In contrast with the previous year, the

chlorophyll-a in the upper basin was significantly greater than that in the lower basin during the growing season in 1999, the first year of fertilization.

The trends in phytoplankton biomass, both with depth and time, do not closely follow the trends in primary productivity. While phytoplanktonic biomass is a function of primary productivity, it is also influenced by several other factors, including predation, parasitism and cell redistribution in the water column due to sinking or turbulence. In addition, high reservoir discharge rates may result in decreased biomass as cells are flushed from the reservoir.

The less favourable spring and summer in 1999 is likely responsible for the overall lower chlorophyll-a in the lower basin during this year. As previously mentioned, there was no significant difference in the chlorophyll-a in the upper basin in 1998 and 1999. Perhaps an increase in chlorophyll-a due to fertilization of this basin in 1999 was sufficient to offset the negative effects of the unfavourable weather during this year.

The mean and range of chlorophyll-a measured in the two basins of Arrow Reservoir in September 1998 and 1999 fall within the same range as those of the four BC reservoirs studied in 1976 (see Table 5.1). The mean chlorophyll-a in the Arrow Reservoir in 1998 and 1999 was only slightly greater than that in 1976. Therefore, there does not appear to be any difference in the mid-September chlorophyll-a 1976 and that in the late 1990's. However, inferences about annual differences in chlorophyll-a can not be made based on the data from a single month.

The chlorophyll-a concentrations in the Arrow Reservoir during the year prior to fertilization were considerably lower than those measured in Kootenay (integrated samples from 0 to 20 m) prior to the initiation of fertilization. From 1975 to 1991, the growing season average chlorophyll-a at station 5 was consistently greater than  $1 \text{ mg/m}^3$  with values ranging from approximately  $1.4 \text{ mg/m}^3$  to greater than  $4 \text{ mg/m}^3$ . During the eight years prior to the initiation of fertilization in this lake (1983-1991) the average growing season chlorophyll-a was approximately  $1.8 \text{ mg/m}^3$ . In the Arrow Reservoir, the seasonal average chlorophyll-a (May to September) during the year prior to fertilization was  $1.45 \text{ mg/m}^3$  in the lower basin and  $1.06 \text{ mg/m}^3$  in the upper basin.

During the first 7 years of fertilization in Kootenay Lake, the growing season average chlorophyll-a was significantly greater than prior to fertilization. The average at

station 5 during this period was  $2.4 \text{ mg/m}^3$ . Chlorophyll-a was also measured at 5 additional stations throughout the fertilization study and the whole lake average chlorophyll-a was estimated to be  $2.7 \text{ mg/m}^3$  (Thompson, 1999).

During the first year of fertilization of the Arrow Reservoir, the seasonal average chlorophyll-a was lower than the previous year in the lower basin ( $0.74 \text{ mg/m}^3$ ) and upper basins ( $0.96 \text{ mg/m}^3$ ). As previously mentioned, it is possible that an increase in chlorophyll-a during the first year of fertilization in the Arrow Reservoir was negated by unusually adverse weather. In addition, the increase in seasonal average chlorophyll-a in Kootenay Lake during the first year of fertilization was relatively small. Since seasonal chlorophyll-a is somewhat variable from year to year in the absence of human intervention, it would be more informative to compare the average seasonal chlorophyll-a for the entire fertilization period to the average for a period prior to fertilization.

For the sake of comparison, in the absence of fertilization, the seasonal average chlorophyll-a in the Arrow Reservoir was even less than that in many extremely oligotrophic coastal lakes. Stockner and Hyatt (1985) report that the average seasonal chlorophyll-a in a wide range of coastal lakes in BC varied from 1 to  $4 \text{ mg/m}^3$ . A comparison of the seasonal average chlorophyll-a of four of these lakes prior to fertilization and during the first year of fertilization yields a large variation in response. The net increase in seasonal average chlorophyll-a in these lakes varied from less than  $0.5 \text{ mg/m}^3$  to greater than  $4 \text{ mg/m}^3$ .

Based on a large quantity of data, Wetzel reports the following general ranges of chlorophyll-a for different trophic categories:  $0.01 - 0.5 \text{ mg/m}^3$  in ultraoligotrophic lakes,  $0.3 - 3 \text{ mg/m}^3$  in oligotrophic lakes,  $2 - 15 \text{ mg/m}^3$  in mesotrophic lakes and  $10 - 500 \text{ mg/m}^3$  in eutrophic lakes. The chlorophyll-a concentrations observed in the Arrow Reservoir during the growing season in 1998 and 1999 fall within the general range reported for oligotrophic lakes.

### 5.1.3 $^{14}\text{C}$ -glucose incorporation

The bacterial activity, as measured by  $^{14}\text{C}$ -glucose incorporation, was highly variable, both with depth and with time. Trends in the data were much less apparent than with the other experiments. In general the bacterial activity increased from the start of

the season to the end with some fluctuation from month to month. In 1999, however, a large peak value occurred in the upper basin in July. No results were obtained for either basin in April and May of 1999. It is likely that the bacterial productivity at this time was lower than the detection limit of this method.

In general there was no apparent relationship between the bacterial activity and abundance. However, it is worth noting that in the upper basin in July 1999 there was a good correlation between bacterial and phytoplankton abundance and activity and fertilization. During this period, bacterial activity and abundance were exceptionally high at 2 m. Both parameters were relatively high at the surface, dropped off at 1 m and peaked at 2 m, reaching the maximum values recorded in both basins in 1998 and 1999. The highest measured primary productivity was also observed in the upper basin in July of 1999 when it peaked at 1 to 2 m. In addition, the summer peak of specific activity in the upper basin was observed during this month.

Statistical analyses of these data indicate that, in each year, there was no significant difference ( $P = 0.99$  and  $0.2$  for 1998 and 1999, respectively) between the bacterial activity in the two basins. In addition, there was no significant difference ( $P = 0.6$ ) in the data in the lower basin in 1998 and 1999. However, at a 95 % confidence level, the bacterial activity in the upper basin was significantly greater in 1999 than in 1998 ( $P = 0.05$ ). It is possible that the increased bacterial activity observed in the upper basin in 1999 is attributable to the fertilization of this basin. Since an increase in primary productivity was observed in the upper basin during fertilization in 1999, it is likely that there was a greater quantity of DOM available to bacteria for degradation.

Wentzell (1987) reports that the glucose uptake rates in Babine Lake in central BC ranged from 100 to 1400 pg glucose/L/hr. The bacterial activity in the Arrow Reservoir was exceptionally small in comparison with that in the oligotrophic Babine Lake. In the Arrow Reservoir, the glucose uptake rates varied from undetectable to 6.75 pg glucose/L/hr during the growing seasons of 1998 and 1999. The glucose uptake rates in the Arrow Reservoir are also a minimum of 2 orders of magnitude smaller than those reported by Hall (1975) for limnocorrals used in a nutrient enrichment experiment in the Bay of Quinte (on the shore of Lake Ontario). In this experiment, the glucose uptake rates (expressed as ug C/L/hr) ranged from approximately 0.7 to 1.2 ug C/L/hr in the



limnocorral enriched with nitrogen and phosphorus and from approximately 0.1 to 0.4 ug C/L/hr in the limnocorral enriched with phosphorus only and the control. When expressed as ug C/L/hr, the glucose uptake rates in the Arrow Reservoir during the growing season in 1998 and 1999 ranged from  $6.7 \times 10^{-5}$  to  $1.9 \times 10^{-3}$ .

Since the algal biomass in the Bay of Quinte limnocorrals (9.16, 28.1 and 25.4 mg chlorophyll-a/m<sup>3</sup> in the control, P enriched and N+P enriched limnocorrals) was much greater than that in the Arrow Reservoir, it is to be expected that the supply of dissolved organic carbon and the bacterial activity would be greater than that in the Arrow Reservoir as well. In Babine Lake, the algal biomass was slightly lower than that in the Arrow Reservoir (0.1 mg chlorophyll-a/m<sup>3</sup>) while the bacterial activity was substantially greater.

Radiolabelled (<sup>3</sup>H) glucose incorporation studies were performed on 8 oligotrophic BC lakes, 4 of which were receiving nutrient additions (MacIsaac et al. 1981). The results of these experiments were reported as glucose turnover times (T), calculated using the equation:  $T = At/U$ , where A = DPM added to each sample, t = incubation time (h) and U = DPM taken up (ie. DPM of a given sample minus DPM of the corresponding blank). The mean annual glucose turnover times (based on monthly sampling from April or May until October or November) in these lakes ranged from 125 to 2200 hours. The turnover times observed in the Arrow Reservoir were substantially greater than those reported by MacIsaac et al. (1981), ranging from 4 300 to 100 000 in 1998 and 3 000 to 39 000 in 1999. It should be noted that in the spring of 1999 no appreciable glucose incorporation was measured. Therefore, the turnover times during these months would be extremely long.

It is likely that the use of a more concentrated <sup>14</sup>C-glucose solution would allow for a more accurate detection of the low bacterial activity in the Arrow Reservoir.

#### 5.1.4 DAPI

The cell abundance in the Arrow Reservoir was estimated using the DAPI technique. In 1998, the monthly weighted average values fluctuated throughout the season with the lowest values recorded in September. In 1999 the lowest monthly weighted average abundance in each basin was in April followed by May. The April

1999 samples were the only ones in the two years where the monthly averages were below  $1 \times 10^6$  cells/ml. In 1999, the bacterial abundance was relatively high from June to August and then dropped off in September. The low bacterial abundance at the end of the season in 1998 and at the start and end of the season in 1999 may be attributable to lower water temperatures (Figure B 9).

In 1998, the cell abundance in the upper basin was slightly greater than that in the lower basin. In the lower basin, there was no significant difference in cell abundance from 1998 to 1999 ( $P = 0.4$ ). However, in the upper basin, the cell abundance was significantly greater in 1999 than in 1998 ( $P = 0.04$ ). There was no significant difference ( $P = 0.4$ ) between the cell abundance in the two basins in 1999. As with the other parameters previously discussed, in the absence of any other factors, any significant changes in bacterial abundance from one year to the next, as a result of annual variations in climate, would be expected to occur to a certain extent in both basins. Since a significant increase in cell abundance from 1998 to 1999 was only observed in the upper basin, it is possible that it is attributable to the initiation of fertilization in this basin in 1999.

Stockner and Hyatt (1984) report seasonal average bacterial abundance data for four extremely oligotrophic coastal BC lakes one year prior to fertilization and during the first year of fertilization. In the absence of fertilization, the seasonal average bacterial abundance in these lakes remained below  $1 \times 10^6$  cells/mL. Prior to fertilization, the bacterial abundance in the Arrow Reservoir was slightly greater than that in these lakes with seasonal averages of  $1.6 \times 10^6$  cells/mL in the lower basin and  $1.4 \times 10^6$  cells/mL in the upper basin. During the first year of fertilization, the seasonal average bacterial abundance in the coastal lakes ranged from slightly less than  $1 \times 10^6$  cells/mL to slightly greater than  $2 \times 10^6$  cells/mL. The average seasonal bacterial abundance in the Arrow Reservoir during the first year of fertilization was comparable to that of the coastal lakes at  $1.7 \times 10^6$  cells/mL in each basin.

Stockner and Shortreed (1989) report seasonal average bacterial abundance data for two lakes in separate biogeoclimatic zones in BC. Both oligotrophic, Quesnel lake is located in the relatively cool, dry interior, while Sproat Lake is located in the relatively warm, moist coastal region. In 1985 and 1986, the bacterial abundance in Quesnel lake

was  $0.97 \times 10^6$  and  $0.42 \times 10^6$  cells/mL, respectively. In Sproat Lake, the bacterial abundance was  $0.72 \times 10^6$  cells/mL in 1985 and  $0.57 \times 10^6$  cells/mL in 1986. Stockner (1987) reports average August-September bacteria numbers in various BC lakes:  $1.45$  and  $0.67 \times 10^6$  cells/mL in fertilized and unfertilized north coastal lakes, respectively,  $1.38$  and  $0.74 \times 10^6$  cells/mL in fertilized and unfertilized south coastal lakes, respectively and  $1.40 \times 10^6$  cells/mL in an interior BC lake. While the bacterial abundance in unfertilized coastal BC lakes was lower than that measured in the Arrow Reservoir, that in fertilized coastal lakes and an interior BC lake were within the range observed in the reservoir.

### 5.1.5 CTC

The percentage of active bacteria in the Arrow Reservoir in 1998 and 1999 ranged from 0.68 to 8.0 %. In 1998, only one month of CTC data is available. Therefore, it is not possible to make a comparison of the bacterial activity in 1998 and 1999 using this method. However, a comparison of the seasonal data in the two basins in 1999 reveals that, in general, the percentage of active bacteria was slightly greater in the upper basin. In each basin the percentage of active cells was substantially greater at the start of the season than at the end.

The CTC incubations were performed in the dark at 4 °C each month. The water temperature was 5 °C at the surface and 4 °C at 10 m in April and 7 °C at the surface and 5 °C at 10 m in May 1999. In early June, the temperature was 11 °C at the surface and 9 °C at 10 m and it remained greater than 10 °C at the surface and 10 m until October. Since the incubation temperature was close to the water temperature during the spring and much cooler than that in the summer, it is possible that the organisms in the summer samples were less tolerant of the cold and remained relatively inactive during the incubation.

The proportion of metabolically active cells in the Arrow Reservoir is relatively low in comparison with that reported by Giorgio and Scarborough (1995) for 24 lakes in southern Quebec, Canada. The percentage of active bacteria in these lakes was estimated using the CTC reduction method with the purpose of assessing the relationship between the proportion of metabolically active bacteria and lake enrichment. The total phosphorus in these lakes ranged from 4.1 to 34.1 ug/L, the total bacterial count ranged

from  $1.88 \times 10^6$  to  $7.50 \times 10^6$  cells/ml and the percentage of active bacteria ranged from 14.1 to 30.7%. The average proportion of active cells in these lakes was 20.9%.

Giorgio and Scarborough also performed a literature review and sorted the percentage of active cells from various studies according to different aquatic environments. They report that the average proportion of active cells was 12% in groundwater, 16% in marine environments, 23% in lakes and 47% in estuaries. In this context, the percentage of active bacteria in the Arrow Reservoir is quite low, indicative of some regulation, such as nutrient availability or lack of a suitable carbon source. While the abundance of bacteria in the reservoir is similar to that of other oligotrophic lakes in BC, the bacterial activity (estimated from  $^{14}\text{C}$ -glucose incorporation rates) is extremely small. These observations are in agreement with the low percentage of active bacteria.

## **5.2 Horizontal transects**

### **5.2.1 Primary productivity**

There were significant monthly differences in the magnitude of primary productivity in the upper basin in 1999. During each of the three months when horizontal transects were performed, the vertical primary productivity profiles were similar to one another and the primary productivity peaked at approximately 2 m during each of these months. Therefore, if the profiles within a given month were similar along the transect of the lake, the transect samples collected at 2 m should provide a good indication of the trends in primary productivity in the transect from month to month and within a given month. It is likely that the vertical profiles were relatively consistent along the transect in a given month as the basin is fairly well mixed and the physical, chemical and biological parameters should be fairly consistent.

The productivity along the transect in August was consistently higher than that in June. In addition, the productivity in June was greater than that in September. These results suggest that the monthly variation in primary productivity observed at the regular sampling location is likely present throughout the transect of the upper basin.

The results of the  $^{14}\text{C}$ -bicarbonate incorporation studies reveal a productivity trend along the north-south transect of the lake. In August and September, the productivity peaked almost half way down the basin, just above Halfway River at station 3. The productivity dropped significantly south of Halfway River and then increased to a second somewhat smaller peak approximately  $\frac{3}{4}$  of the way down the transect of the basin. The productivity then decreased to the monthly minimum value in the southern end of the basin, adjacent to Nakusp. It is highly likely that the drop in productivity just below Halfway River is due to the influence of this inflow.

In June, the transect productivity peaked just below Halfway River at station 4. No samples were processed from station 3 during this month. Since no drop in productivity is observed downstream of Halfway River during this month, it is possible that the difference in temperature between the tributary and the lake is large enough that the colder, denser tributary water dives below the epilimnion upon entering the lake.

It appears that the maximum effects of fertilization on primary productivity occur approximately mid-way down the upper basin. This parameter increased downstream of the location of fertilizer addition, peaked approximately halfway down the upper basin and then declined as the additional nutrients became bound up in the biomass. Ideally, at least two vertical profiles should be performed at different locations along the horizontal transect to assess the degree of uniformity of these profiles. Alternatively, 2 to 3 primary productivity incubations could be performed at different depths at 3 or more locations along the horizontal transect.

### **5.2.2 Chlorophyll-a**

The vertical chlorophyll-a profiles in the upper basin in 1999 changed significantly from month to month. In June, a relatively sharp peak in chlorophyll-a was observed at 2 m. This peak was one of the highest observed in the reservoir and it was followed by a rapid decline in chlorophyll-a with depth. In August and September, the chlorophyll-a at 2 m was less than half of that observed at 2 m in June and the peaks were more broad during these two months with maximum values at 15 and 10 m, respectively. Therefore, a month to month comparison of the horizontal transect samples is meaningless since these samples, which were all collected at 2 m, would be

representative of different regions of the vertical profile. In addition, since the shapes of the profiles (sharp peak at 2 m in June and broad plateau from approximately 5 m to at least 15 m in August and September) were different, a comparison of the depth of maximum algal biomass each month would not provide a reliable indication of the monthly differences in the total algal biomass at a given location.

Within a given month, there was some variability in the algal biomass from station to station. The chlorophyll-a at station 3.5, which is actually the chlorophyll-a at 2 m from the vertical profile, conforms relatively well with the other stations in August and September. In June, the chlorophyll-a at station 3.5 was high relative to the other stations.

Within a given month, the horizontal transect samples may be compared to note any trends, assuming that the vertical profiles are consistent along the transect. As with the vertical profiles for primary productivity, it is likely that those for chlorophyll-a are similar along the transect in a given month. The degree of uniformity of algal biomass along the transect may be assessed by sampling several depths at 2 or more locations along the transect.

### **5.3 Containment experiments**

The N:P ratio of the fertilizer changed over the season as did the concentration of phosphorus. The seasonal phosphorus additions were designed to mimic the natural phosphorus loading, with the maximum additions coinciding with spring freshet. The phosphorus loading increased from mid-April to the peak at the end of May and then decreased to a constant value, which was maintained from mid-June to mid-August. The loading was then further reduced in mid-August and maintained at a low value until fertilization was terminated in early September. The N:P ratio of the fertilizer increased incrementally every 3 to 4 weeks throughout the season starting at 0.67:1 in April and peaking at 7.5:1 in late August. The rationale for the increasing N:P ratio was to discourage the growth of nitrogen-fixing cyanobacteria (blue-green algae), a nuisance species which often dominates the phytoplankton community when the N:P ratio is low (Stockner and Shortreed 1988).

The purpose of the containment experiments was to determine whether the N:P ratio of the fertilizer at a given time was optimal for primary production. The results indicate that, in July, with the actual N:P ratio of the fertilizer (6:1), a significant improvement in phytoplankton biomass and productivity should be observed over the lower N:P ratio (4.5:1). In addition, no significant increase in these parameters should be observed with a higher N:P ratio (7:1). However, since the N:P ratio changes throughout the fertilization period, generalizations about the N:P ratio can not be made based on the results of a single experiment. Ideally, monthly containment experiments, using the improved method designed in July, should be performed throughout the seasonal fertilization period. In addition, the containment experiments should be further modified to account for phytoplankton adaptation. It has been previously documented (Stockner and Antia, 1976) that an exposure period of up to 20 days or more may be required to allow for algal adaptation to the test conditions.

## **5.4 Stream Bioassays**

### **5.4.1 Full Bioassays**

The concentrations of both nitrogen and phosphorus in each of the 8 samples appear to be limiting growth. The net growth was substantially greater with the addition of a combination of these nutrients than with all other treatments, including the addition of one of the two nutrients alone. Samples collected from Inonoaklin Creek, which receives nutrient inputs from local farms, appear to be limited by nitrogen. Results from samples from Mosquito Creek, which had virtually no nitrate nitrogen in September compared to 89 mg/L in May, indicate that this creek was nitrogen limited in September. The remainder of the samples appear to be nitrogen and phosphorus co-limited.

Since there was little difference in algal growth in filtered and unfiltered samples, it is unlikely that particle bound nutrients stimulated growth over the bioassay period. Due to the short-term nature of the bioassay and the affinity of phosphorus for particulate matter, it is possible that some stimulation would be observed over a longer period of time. In addition, a longer exposure to the low nutrient concentrations may stimulate tolerance for these conditions in the *S. capricornutum* population, which was cultured in

a nutrient rich broth. Stockner and Antia (1976) report that 4 day bioassays are not sufficiently long to take into account adaptations of the test organism to the test conditions. The natural populations of periphyton in the streams are likely better adapted to the low nutrient concentrations than the *S. capricornutum* cultures used in the bioassays.

#### 5.4.2 Modified Bioassays

A range in net growth was observed in samples spiked with a combination of nitrogen and phosphorus. Since the samples were incubated under saturating light, the difference in net growth in samples spiked with N and P is not due to differential light limitation resulting from different levels of turbidity. However, different levels of turbidity may contribute to the differences in net growth due to the adsorption of the nutrients to the particles. This can not be checked since no turbidity analyses were performed with these samples. Another potential source of the differences in net growth in these samples is the presence of natural growth inhibiting substances in various quantities (Paul Harrison, personal communication). It appears that the spread of net growth in samples enriched with N and P is partially the result of a combination of these factors. The net growth in sample alone is likely affected by the same factors.



## 6. SUMMARY AND CONCLUSIONS

### 6.1 Vertical transects

The results of the vertical transects may be summarized as follows:

- The primary productivity in the upper basin during the first year of fertilization (1999) was greater than that in the upper basin during the year prior to fertilization (1998). In the lower basin there was no significant difference between the primary productivity in 1998 and 1999. In 1999, the primary productivity in the upper basin, which was receiving fertilizer, was greater than that in the lower basin. In 1998, prior to fertilization, there was no significant difference in the primary productivity of the two basins.
- There was no significant difference in the chlorophyll-a in the upper basin during the first year of fertilization and the year prior to that. In contrast, the chlorophyll-a in the lower basin was significantly lower in 1999 than in 1998. During the first year of fertilization, the chlorophyll-a in the fertilized upper basin was greater than that in the lower basin. However, during the year prior to fertilization, the chlorophyll-a was not significantly different in the two basins.
- The microbial activity did not differ significantly in the two basins during the pre-treatment year, nor was there any significant difference during the first year of fertilization. In the lower basin, the microbial activity was not significantly different in 1998 and in 1999. In the upper basin, however, the microbial activity was greater during the first year of fertilization than the year prior to that.
- As with the microbial activity, the bacterial abundance did not differ significantly in the two basins of the Arrow Reservoir both prior to and during the first year of fertilization. Also, while the difference in bacterial abundance prior to and during fertilization was significant in the upper basin, it was not significant in the lower basin.

Natural annual differences in the seasonal productivity and biomass of phytoplankton and bacteria would be anticipated, especially when comparing a warm, dry year such as 1998 with a cool, wet year such as 1999. In successive contrasting years such as these, it is likely that productivity and biomass would be lower during the cooler, wetter year, i.e. 1999.

The results of the vertical transects indicate that the productivity and biomass of phytoplankton and bacteria in the upper basin was enhanced during the first year of fertilization. While primary productivity and bacterial activity and abundance were not significantly different from one year to the next in the lower basin, these parameters were all greater in 1999, during fertilization, than in 1998 in the upper basin. Algal biomass in the upper basin was not significantly greater during the first year of fertilization than during the year prior. However, the algal biomass in the lower basin was significantly lower during the first year of fertilization than during the pre-treatment year. It appears that, in the upper basin in 1999, fertilization has cancelled the negative effects of the poor climate on algal biomass seen in the lower basin.

It is recommended that further studies of the algal and microbial communities be performed annually throughout the fertilization project. This would help discriminate natural annual differences from those attributable to fertilization. In addition, annual studies throughout the fertilization period would provide insight into the long term effects of fertilization on the algal and microbial communities.

## **6.2 Horizontal transects**

The results of the horizontal transects may be summarized as follows:

- The monthly differences in primary productivity in 1999 were observed throughout the north-south transect of the upper basin at the transect depth of 2 m.
- There was a trend in the primary productivity along the transect in 1999, in which the maximum productivity just north of Halfway River is followed by a local minimum and a second smaller peak in productivity further south, closer to Nakusp.

- The monthly differences in phytoplankton biomass in 1999 were not observed throughout the north-south transect of the upper basin. However, since the monthly chlorophyll-a vertical profiles are drastically different, month to month comparisons of samples collected at 2 m are inappropriate.
- There did not appear to be a trend in the phytoplankton biomass along the transect in 1999.

Assuming that the vertical profile is relatively uniform along the horizontal transect, the monthly differences in primary productivity are consistent throughout the transect. Therefore, the primary productivity results from the vertical profile provide an adequate representation of the monthly changes in this parameter in the pelagic region of the upper basin. In order to confirm the uniformity of the primary productivity vertical profile along the transect, primary productivity incubations should be performed at additional depths at one or more stations.

### **6.3 Containment Experiments**

The results of the containment experiments indicate that, in comparison with N:P ratios of 4.5:1 and 7:1, the N:P ratio of the fertilizer in July (6:1) is optimal for primary productivity. It would be beneficial to perform the modified containment experiments at several times during a season of fertilization to determine the efficiency of the changing N:P ratio. In addition, the incubation period for the carboys should be extended to at least 20 days to allow for phytoplankton acclimation. This is probably an unreasonable time for the small 20 L carboys due to concerns about periphyton growth. Therefore, larger microcosms should be utilized for the containment experiments.

### **6.4 Stream Bioassays**

The results of the stream bioassays are as follows:

- The concentrations of biologically available nitrogen and phosphorus are so low as to be growth-limiting to phytoplankton and attached algae in Arrow Reservoir.
- Algal growth in samples from the Inonoaklin Creek, which is influenced by local agricultural activities, was limited by nitrogen to a greater extent than phosphorus.
- The maximum potential short term net growth varied significantly in the different samples. This was likely due to differences in turbidity and the presence of growth inhibiting substances.

It would be informative to modify this bioassay such that the natural algal community is used to assess the nutrient availability and growth potential of these samples. An incubation period of at least 20 days should also be considered to allow for phytoplankton acclimation.

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## **APPENDIX A**

## SAMPLE CALCULATIONS

### Primary Productivity

Data: light bottle DPM = A  
 dark bottle DPM = B  
 alkalinity = alk (mg/L as CaCO<sub>3</sub>)  
 activity of radioisotope solution = DPM/ml  
 Bottle volume (V) = 300 mL  
 Incubation time (t) = 4 hours

$$\text{net DPM} = A - B$$

$$\text{primary productivity (mg C/L/hr)} = \frac{\text{net DPM}}{\text{DPM/mL}} * \text{alk} * \frac{12 \text{ mg C}}{100 \text{ mg CaCO}_3} * \frac{1000 \text{ mL/L}}{V} * \frac{1}{t}$$

### Chlorophyll-a

Data: Initial reading = A  
 Acidified reading = B  
 Volume of acetone extract (v) = 10 mL  
 Volume of sample filtered (V) = 500 mL  
 Acid ratio correction factor = 1.96  
 Slope of calibration curve = 0.729

$$\text{Chlorophyll-a (mg/m}^3\text{)} = 2.069 * 0.994 * (A-B) * v/V$$

### <sup>14</sup>C-glucose incorporation

Data: sample syringe DPM = A  
 Control syringe DPM = B  
 Activity of radioisotope solution = DPM/mL  
 Specific activity (sp. act.) = 286.9 mCi/mmol  
 Molecular Weight of glucose (MW) = 180 g/mol  
 1 uCi = 2.2 x 10<sup>6</sup> dpm  
 sample volume (V) = 18 mL  
 incubation time (t) = 3 hours

$$\text{net DPM} = A - B$$

$$\text{available glucose (ug)} = \frac{\text{MW}}{\text{sp. act.}} * \frac{\text{DPM/mL}}{2.2 \times 10^6 \text{ DPM/uCi}}$$

$$\text{Bacterial activity (ug glucose/L/hr)} = \frac{\text{net DPM}}{\text{DPM/mL}} * \text{available glucose} * \frac{1000 \text{ mL/L}}{V} * \frac{1}{t}$$

### DAPI

Data: Total filter counts = cells  
 Total grids counted = grids  
 Volume of filtered sample (V mL) = 10 mL  
 Area of grid = 97 um x 97 um  
 Area of filter = 490.874 mm<sup>2</sup>  
 Areal microscope conversion factor (CF) = 52170.6

$$\text{Mean field count} = \frac{\text{cells}}{\text{grids}} * 100 \text{ grids/field}$$

$$\text{Active cells/ml} = \text{mean field count} * \frac{CF}{V}$$

### CTC

Data: Total filter counts = cells  
 Total grids counted = grids  
 Area of grid = 97 um x 97 um  
 Area of filter = 490.874 mm<sup>2</sup>  
 Volume of filtered sample (V mL) = 10 mL  
 Areal microscope conversion factor (CF) = 52170.6

$$\text{Mean field count} = \frac{\text{cells}}{\text{grids}} * 100 \text{ grids/field}$$

$$\text{Active cells/ml} = \text{mean field count} * \frac{CF}{V}$$

$$\% \text{ active bacteria} = \frac{\text{active cells/ml}}{\text{total cells/ml}} * 100 \%$$

### Alkalinity

Data: Initial burette reading = A  
 Final burette reading = B  
 Normality of H<sub>2</sub>SO<sub>4</sub> titrant (N) = 0.02  
 Volume of sample titrated (V) = 100 mL

Volume of acid = B-A

$$\text{Alkalinity (mg/L as CaCO}_3\text{)} = \frac{\text{volume of acid} * N * 50,000}{V}$$

### Depth Weighted Average

Weighted average of parameter “x” from the surface to a depth of 15 m, where parameter “x” was measured at the surface (0 m) and depths 1, 2, 5, 10 and 15 m:

X<sub>n</sub> = value of parameter “x” at depth “n”

Since intervals between sampling depths are not equal, each X<sub>n</sub> is multiplied by a weighting factor prior to calculating the average.

Weighting factor for X<sub>n</sub> =

$$(\text{interval between } X_{n-1} \text{ and } X_n)/2 + (\text{interval between } X_n \text{ and } X_{n+1})/2$$

See figure \*\* for further details.

$$\text{weighted average} = \frac{(0.5 * X_0) + (1 * X_1) + (2 * X_2) + (4 * X_5) + (5 * X_{10}) + (2.5 * X_{15})}{15}$$

The units are the same as those for X<sub>n</sub>.

### Daily Areal Primary Productivity

P<sub>n</sub> = estimated daily primary productivity (mg C/m<sup>3</sup>/hour) at depth “n”

Estimated daily primary productivity from each depth is multiplied by the weighting factor described in the depth weighted average section above to estimate the areal primary productivity in mg C/m<sup>2</sup>/hour:

$$\text{Areal productivity} = (0.5 * P_0) + (1 * P_1) + (2 * P_2) + (4 * P_5) + (5 * P_{10}) + (2.5 * P_{15})$$

Figure A 1 - Depiction of the determination of weighting factors for the calculation of weighted average

| <u>Depth</u> | <u>Intervals<br/>between<br/>depths</u> | <u>Weighting<br/>factor</u> |
|--------------|---|-----------------------------|
| 0            |   | 0.5 m                       |
| 1            | 1 m                                     | -----                       |
| 2            | 2 m                                     | -----                       |
| 5            |   | 2 m                         |
|              | 3 m                                     | -----                       |
|              |   | 4 m                         |
|              | 5 m                                     | -----                       |
| 10           |   | 5 m                         |
|              | 5 m                                     | -----                       |
|              |   | 2.5 m                       |
| 15           |   | -----                       |



## **APPENDIX B**

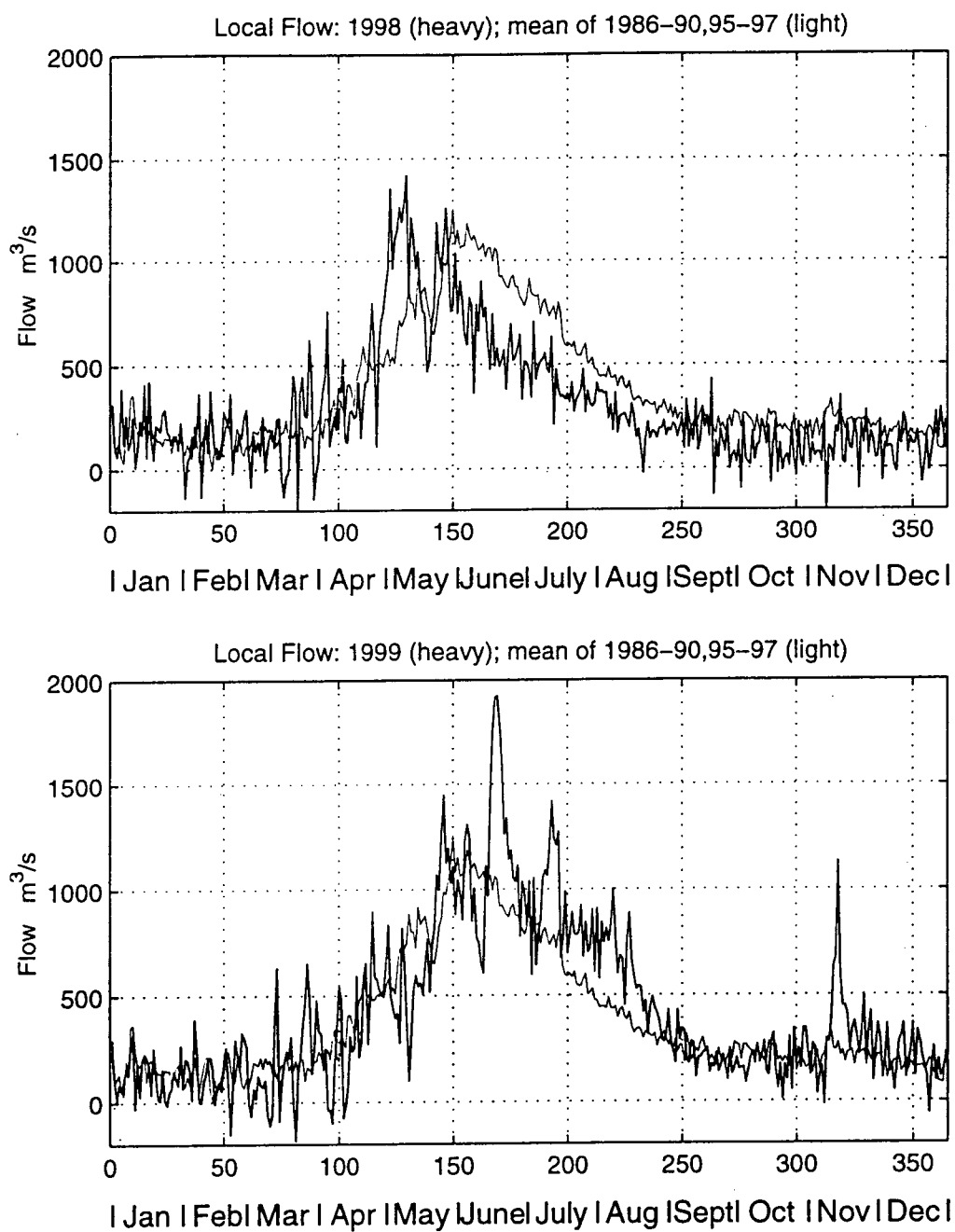


Figure B.1 – A comparison of local flow profiles for the Arrow Reservoir in 1998 and 1999 with the historical average

Figure B 2 - 1998 light profiles for the upper basin of the Arrow Reservoir

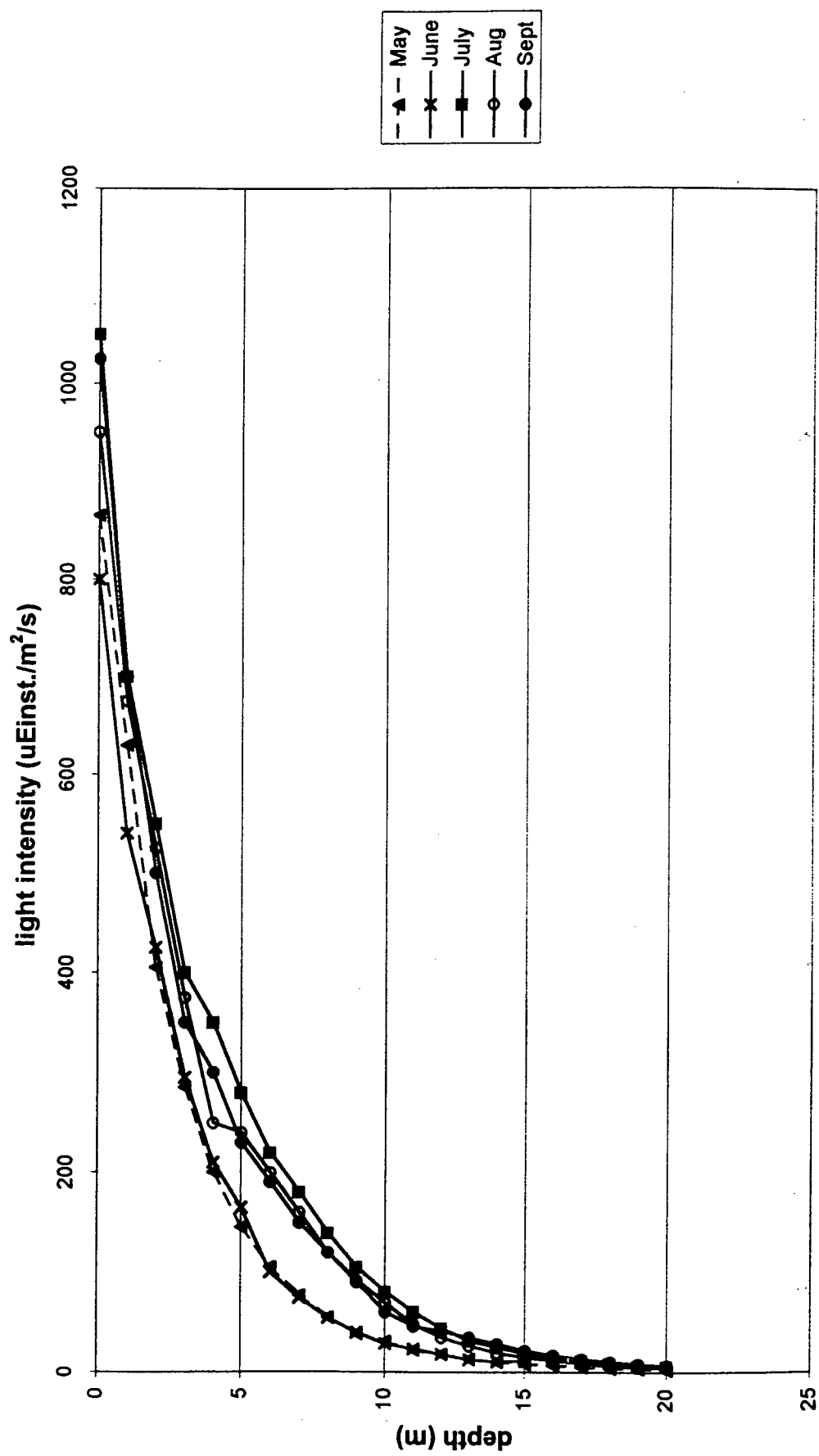


Figure B 3 - 1998 light profiles for the lower basin of the Arrow Reservoir

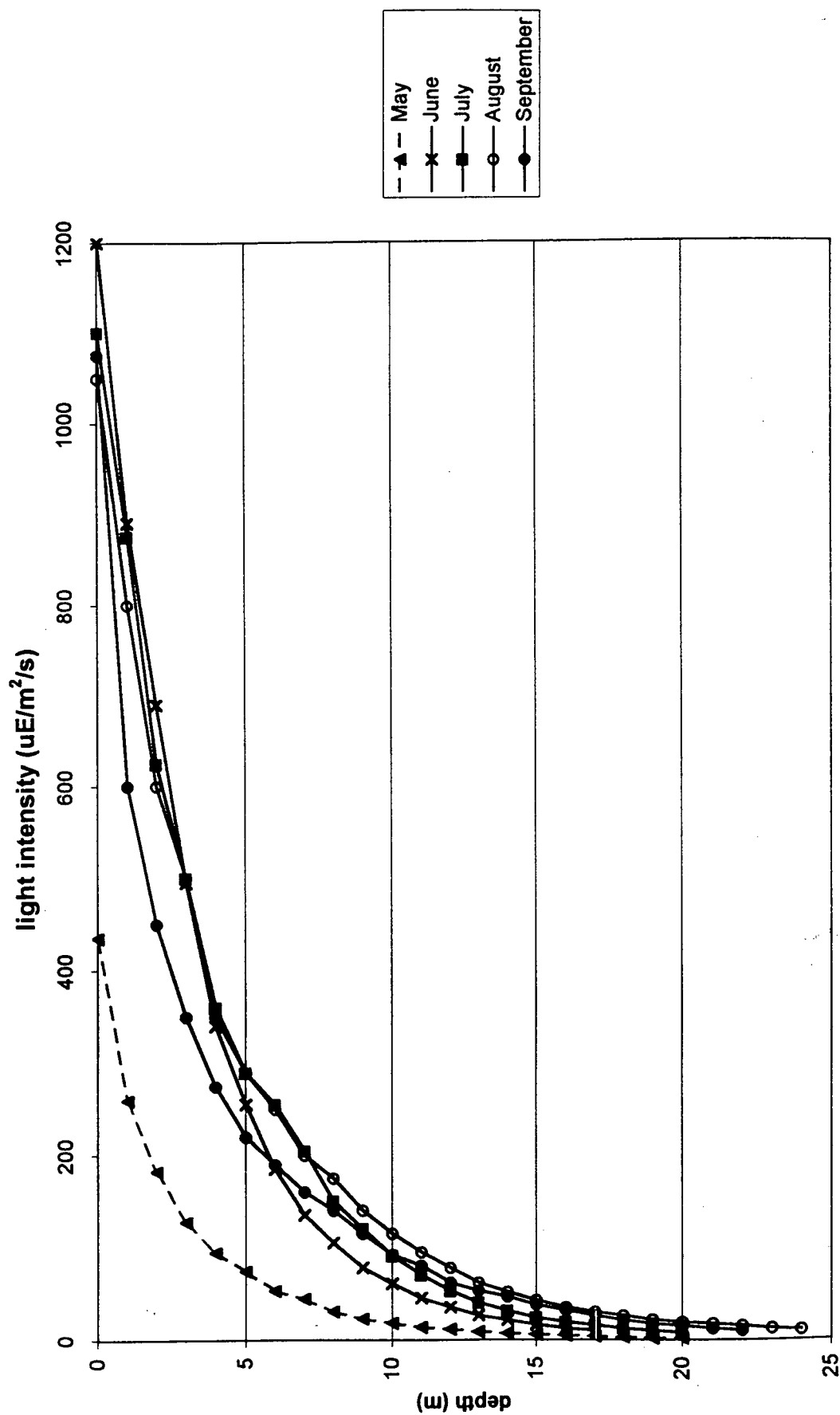


Figure B 4 - 1999 light profiles for the upper basin of the Arrow Reservoir

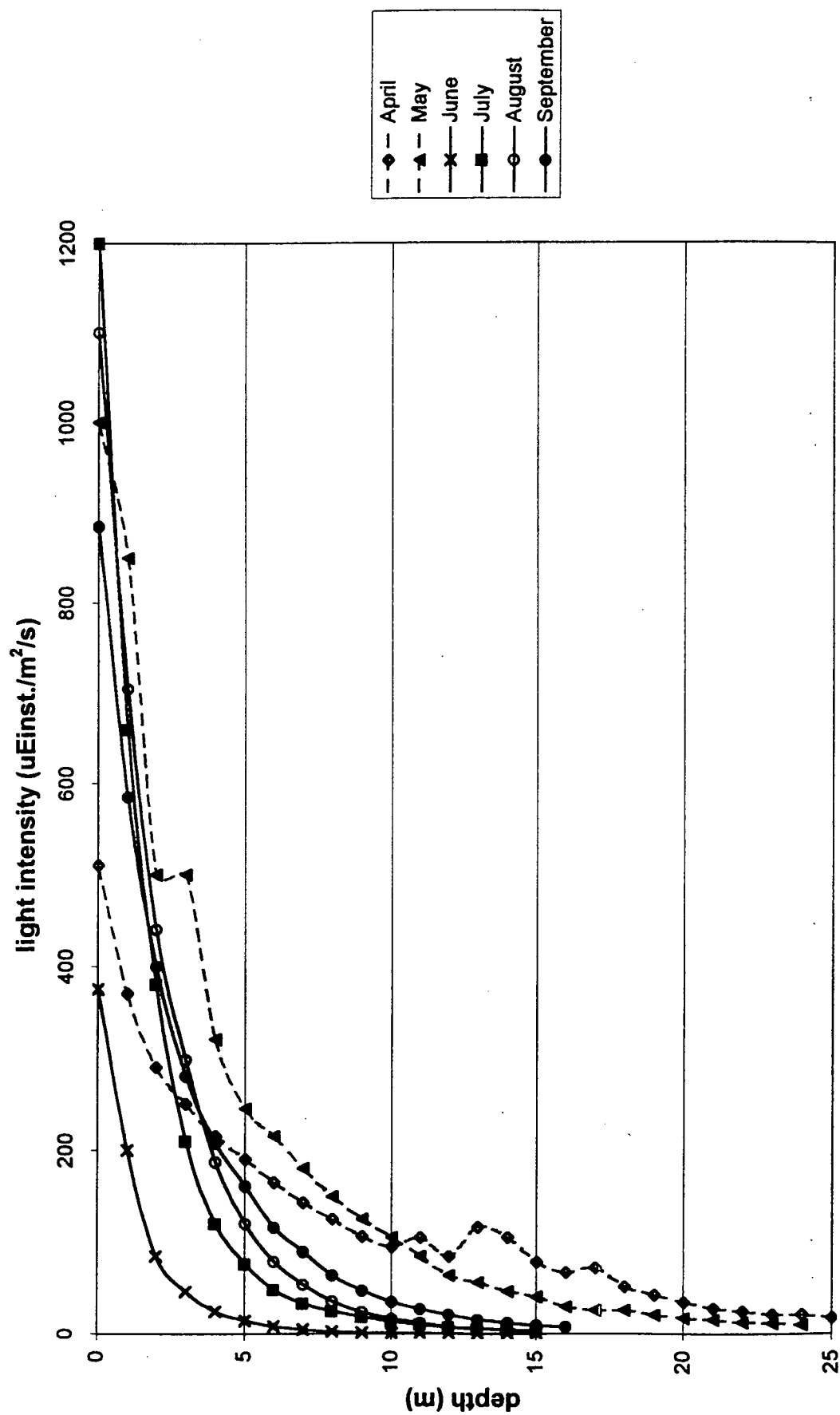
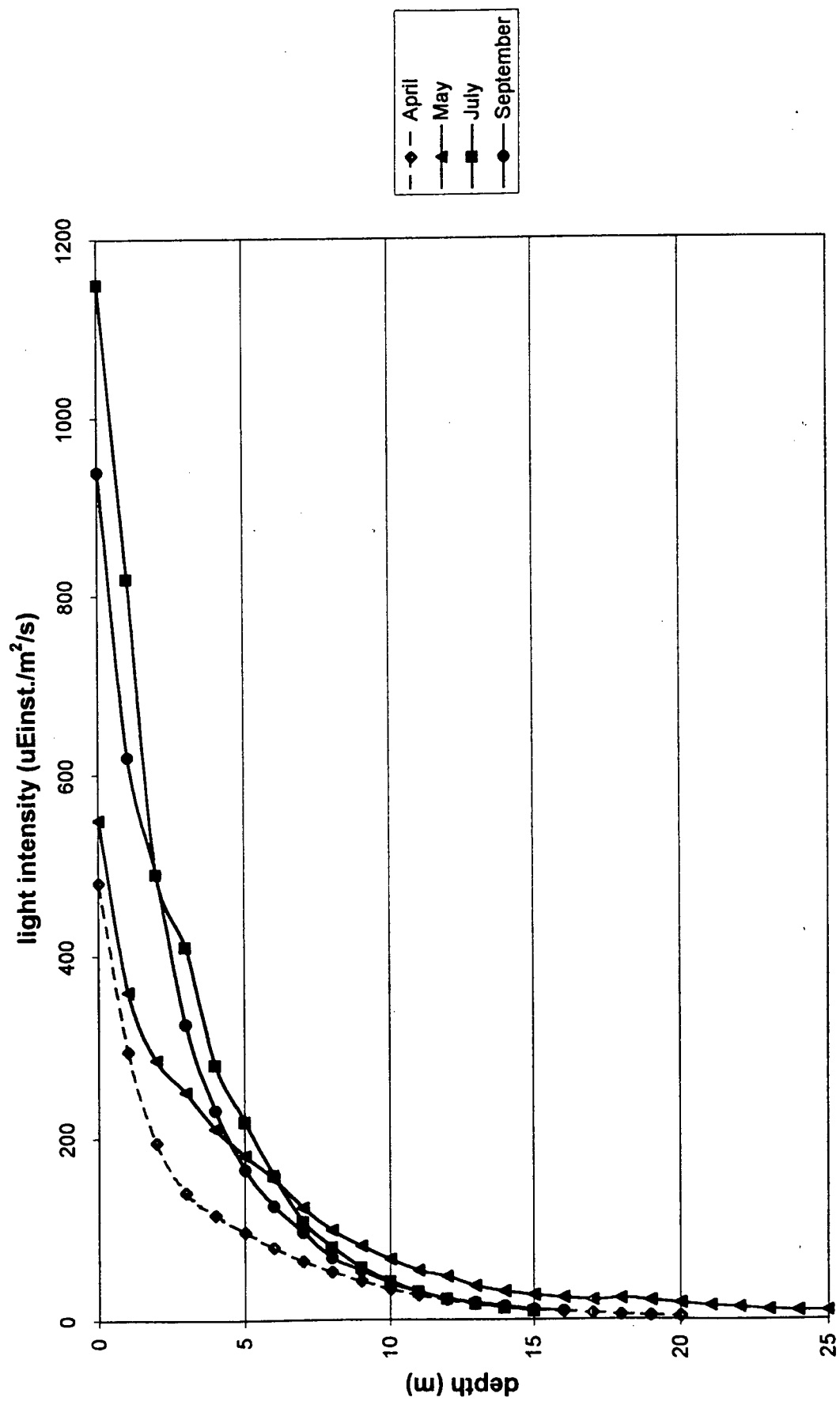


Figure B 5 - 1999 light profiles for the lower basin of the Arrow Reservoir



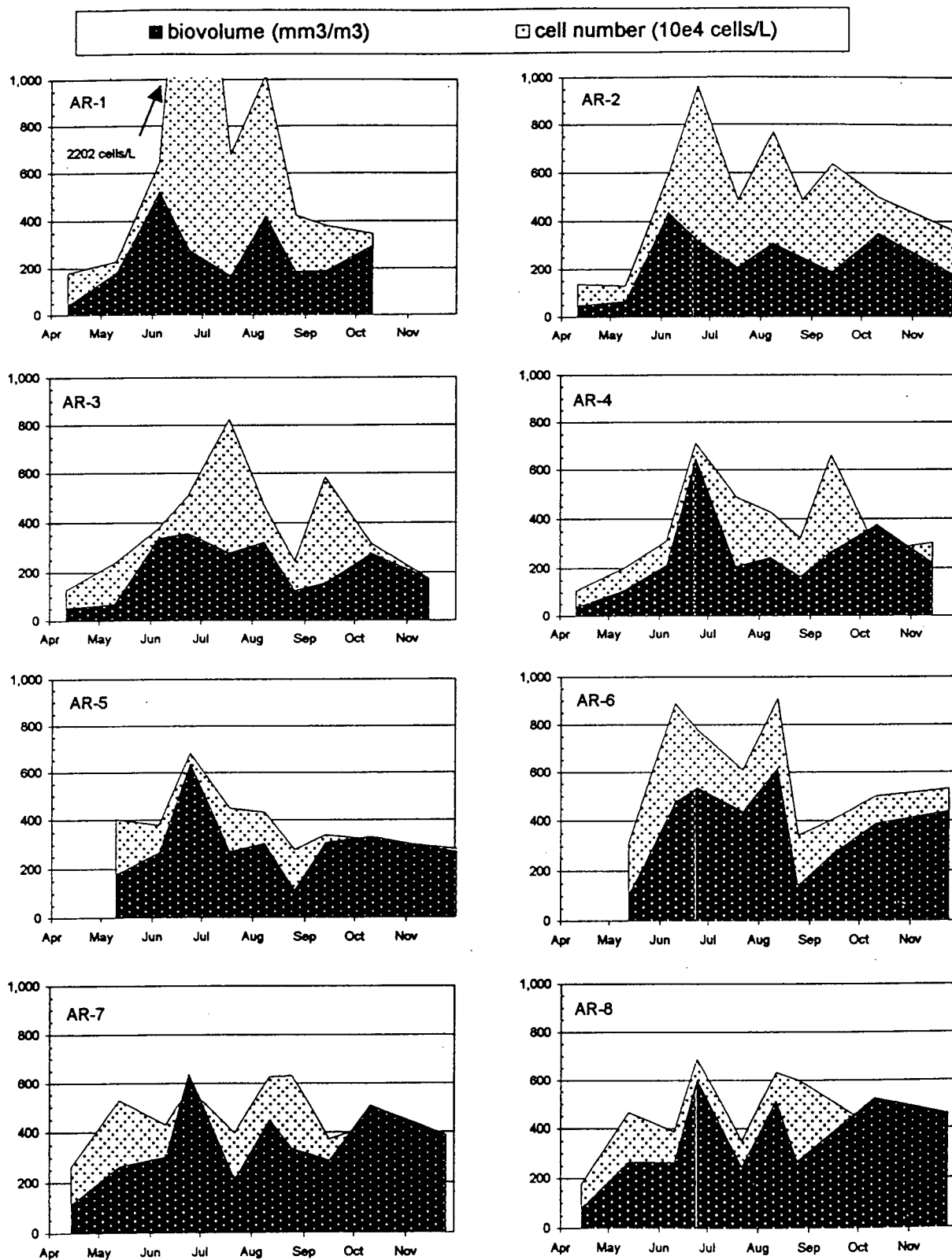
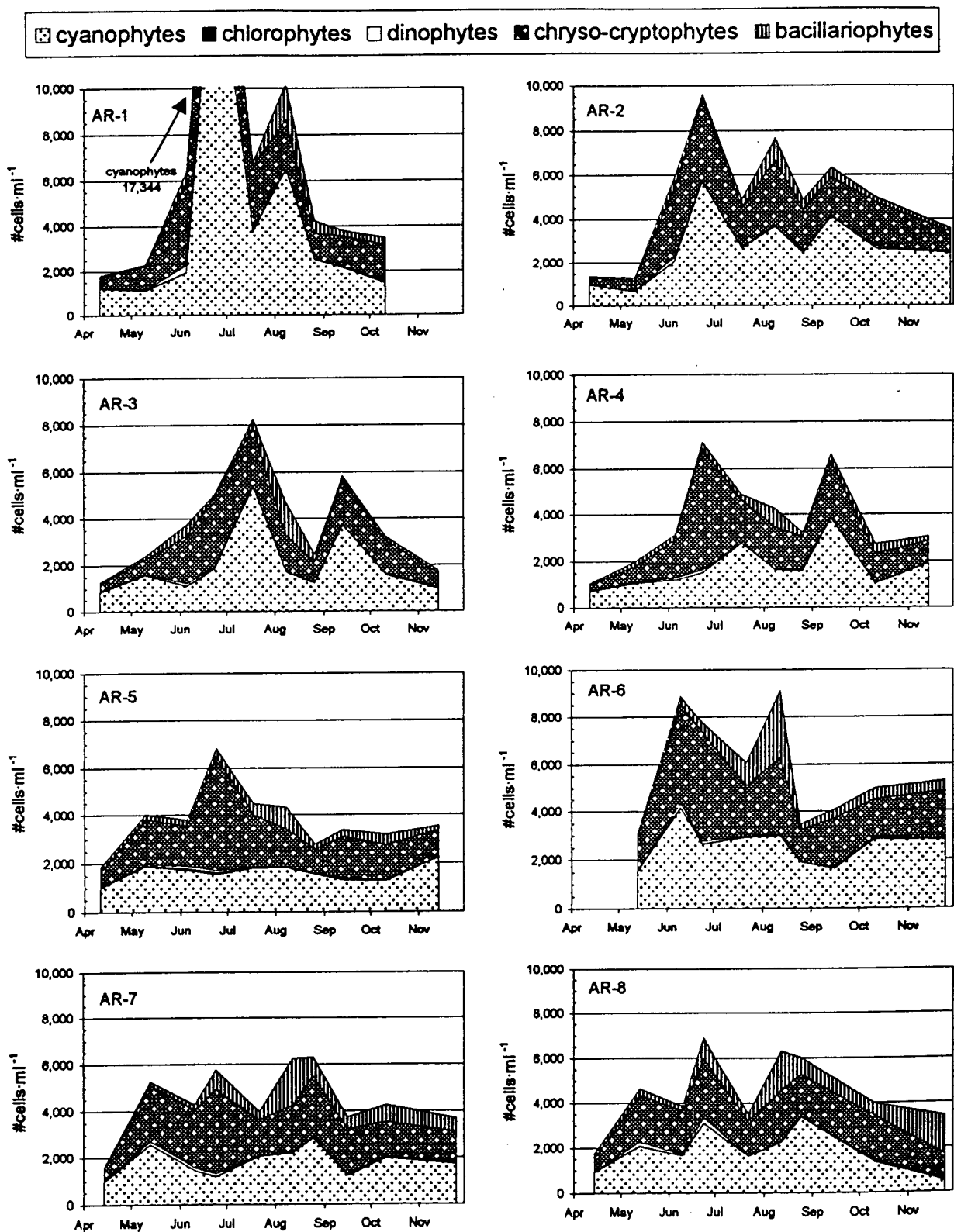
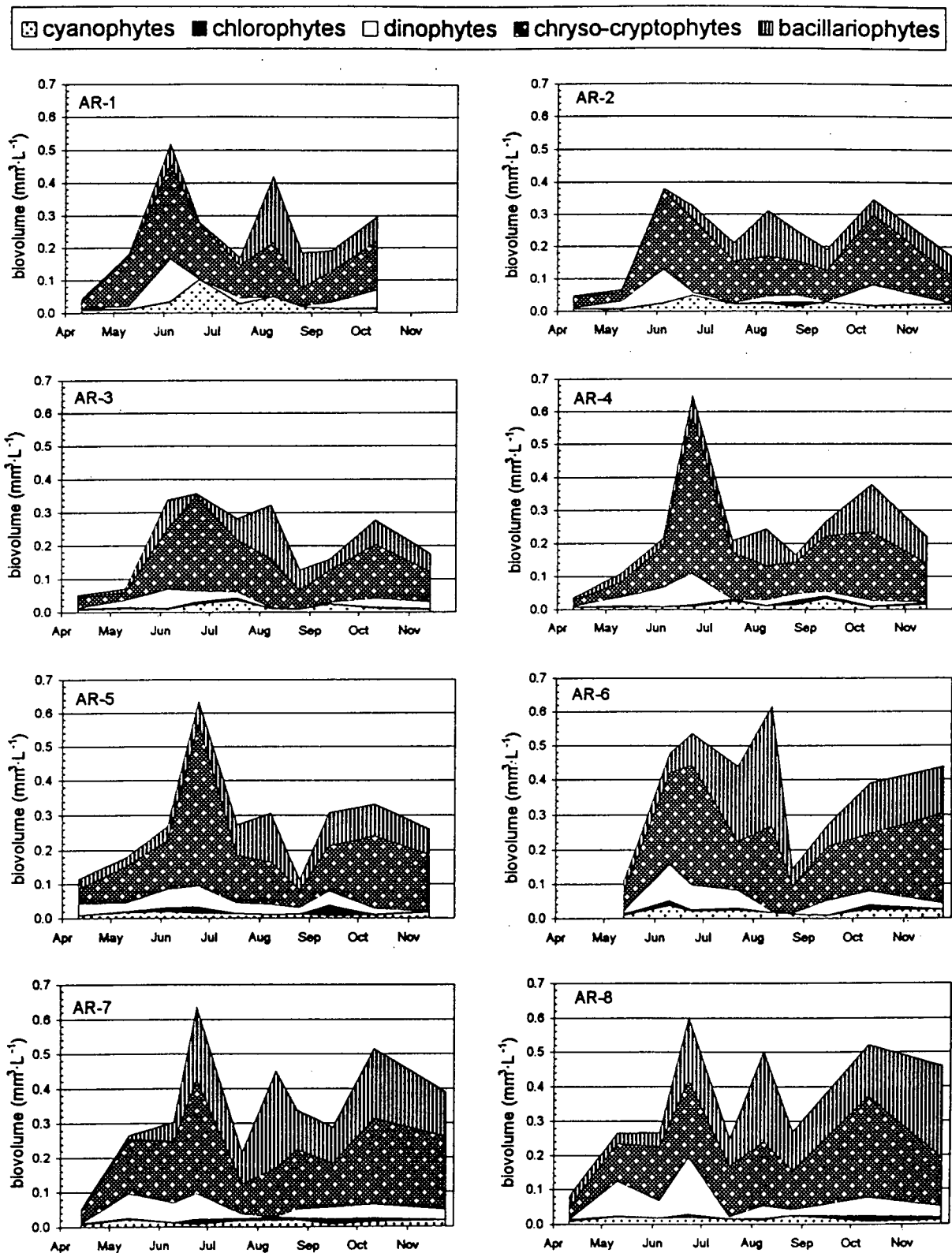


Figure B6 Seasonal (May-Oct.) epilimnetic phytoplankton abundance and biomass in Arrow Reservoir in 1999.



**Figure B7** Seasonal (May-Oct.) abundance of the major phytoplankton classes in Arrow Reservoir in 1999.





**Figure B8** Seasonal (May-Oct.) biomass of the major phytoplankton classes in the Arrow Reservoir in 1999.

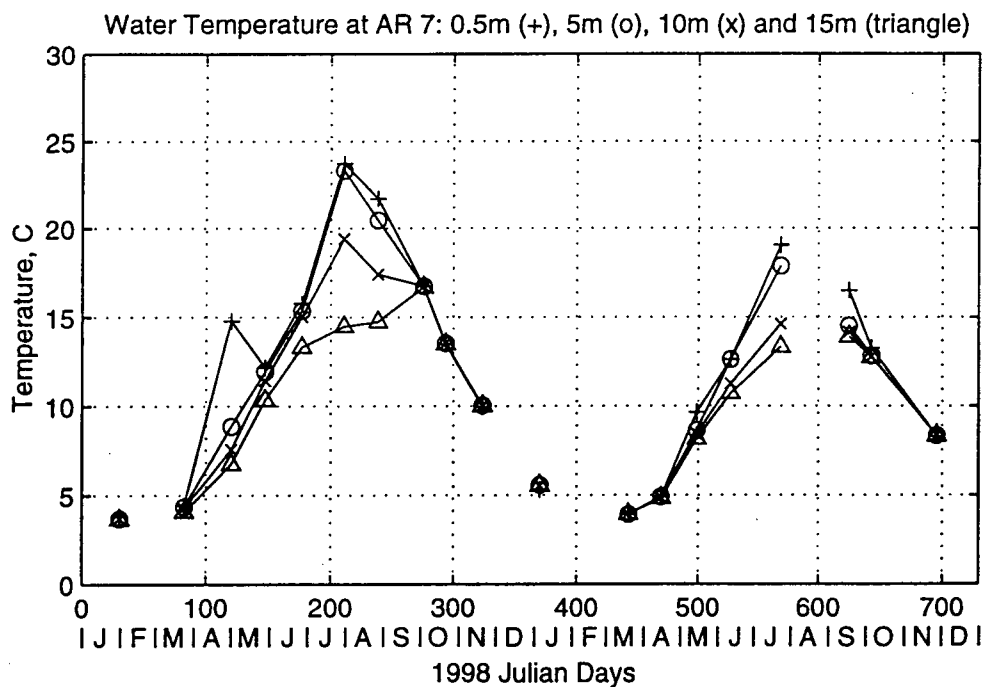
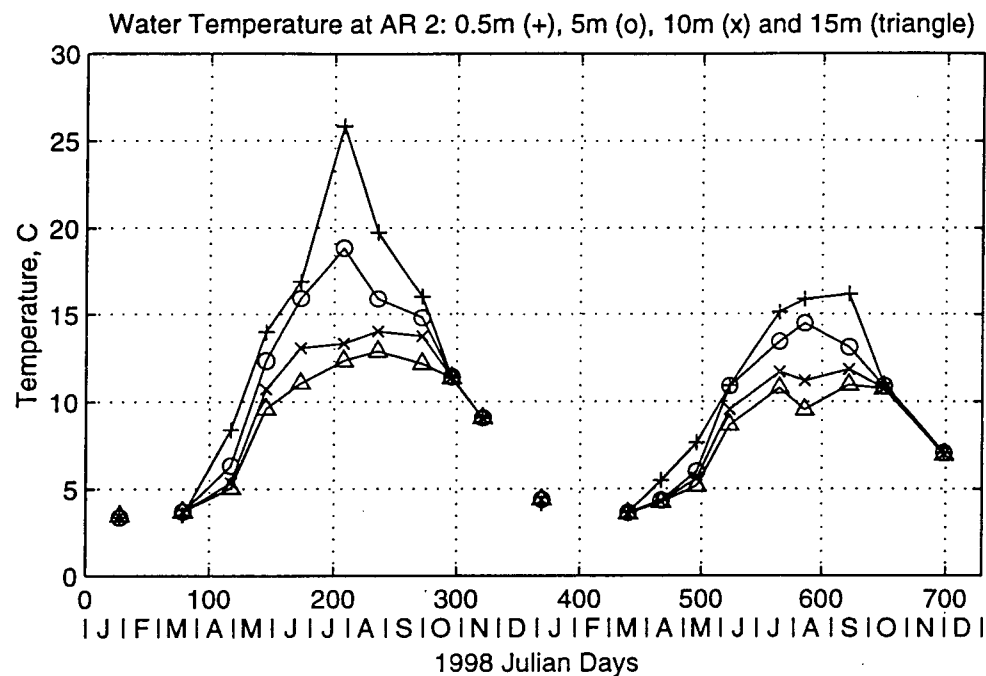


Figure B.9 – Seasonal profiles of the near surface water temperature in the Arrow Reservoir in 1997, 1998 and 1999

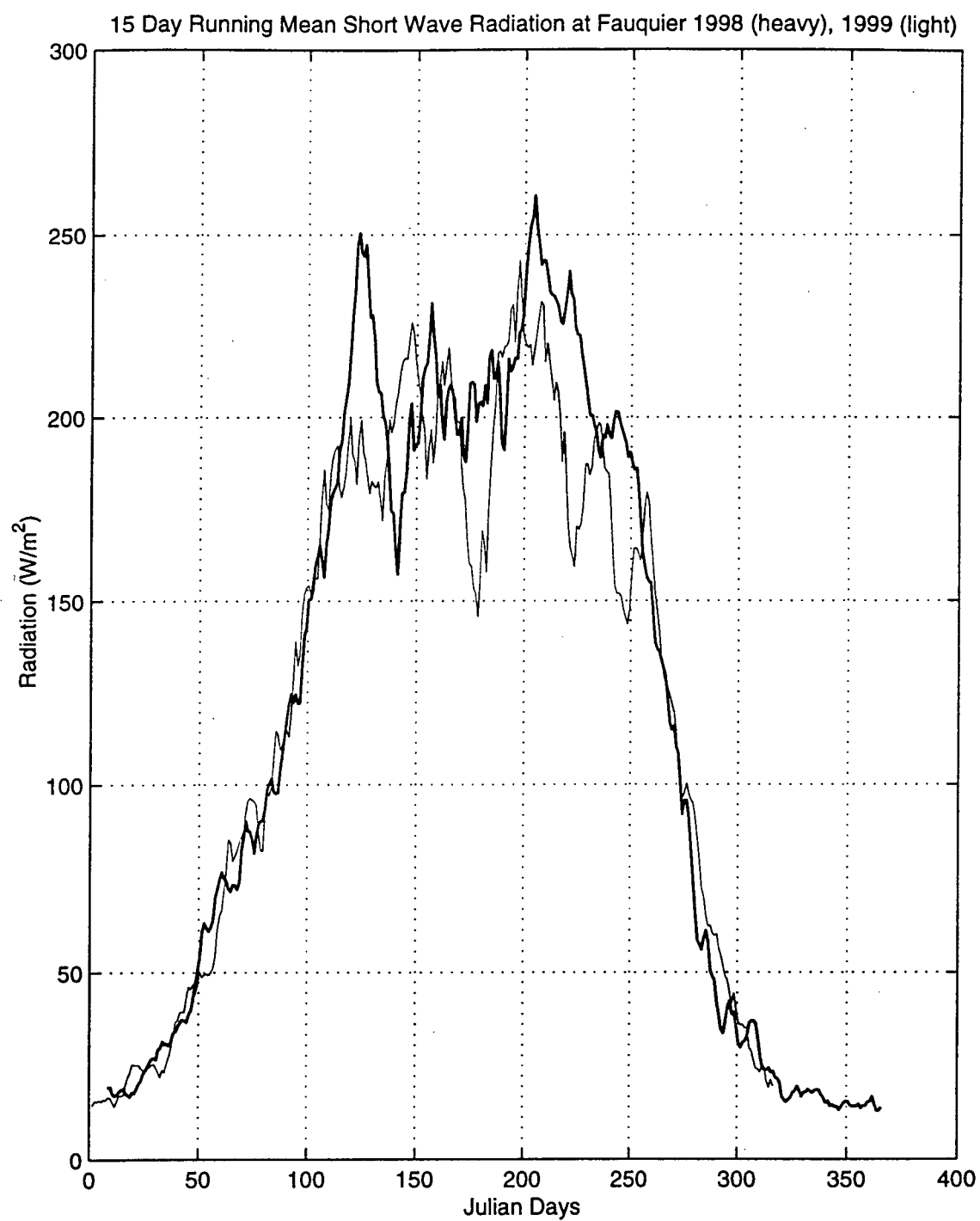


Figure B.10 – Seasonal profiles of the short wave solar radiation in Fauquier in 1998 and 1999

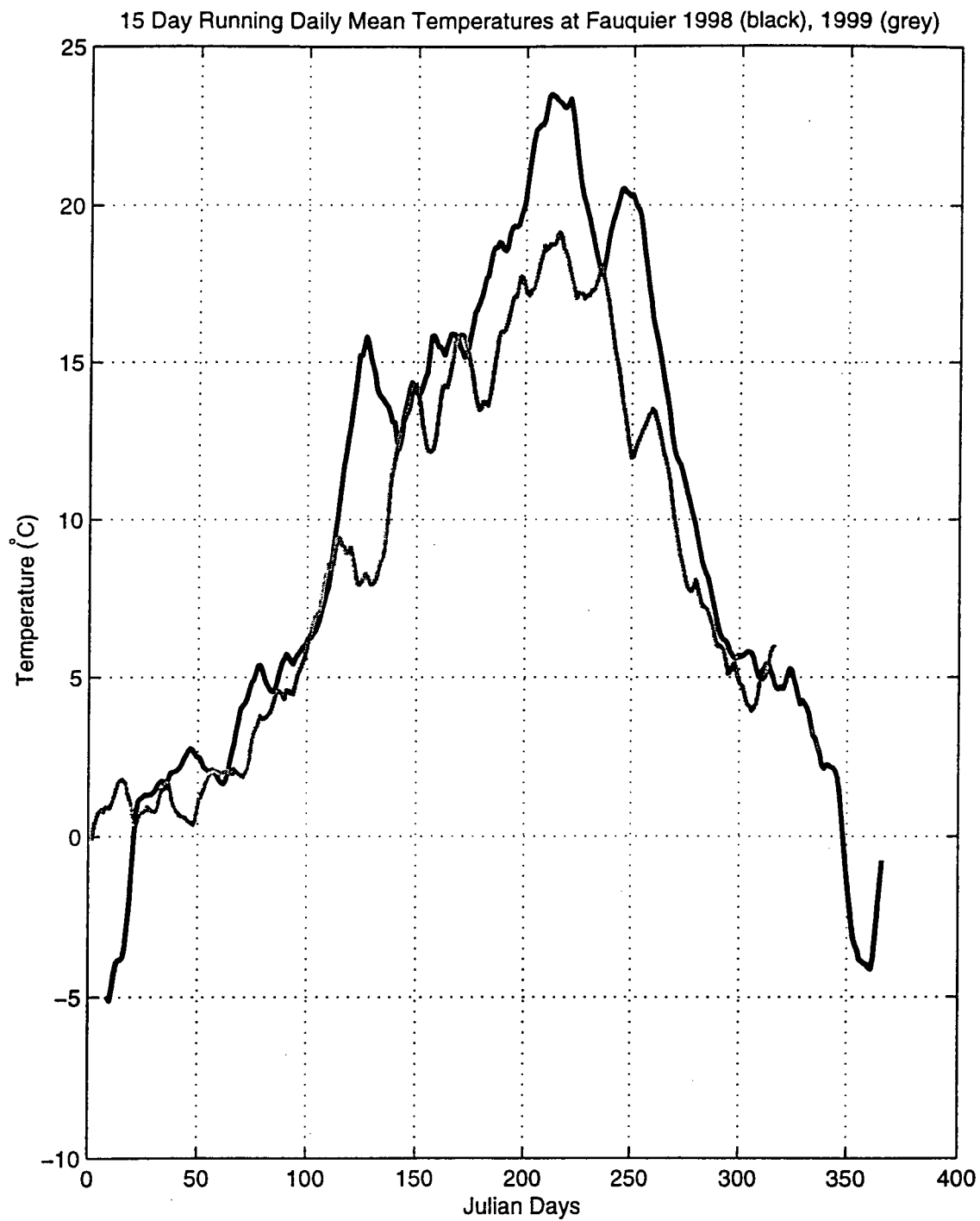


Figure B.11 – Seasonal air temperature profiles for Fauquier in 1998 and 1999