CONCENTRATION, DISTRIBUTION, AND BACTERIAL COLONIZATION OF TRANSPARENT EXPOLYMER PARTICLES (TEP) AMONG MACKENZIE DELTA LAKES

by

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B.Sc. (Honours), University of Waterloo, 2004

THESIS SUBMITTED IN PARTIAL FULFILLMENT OF
THE REQUIREMENTS FOR THE DEGREE OF

MASTER OF SCIENCE

In the
Department of Geography

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SIMON FRASER UNIVERSITY

Fall 2008

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ABSTRACT

Concentrations, distributions, and bacterial colonization of transparent exopolymer particles (TEP) were tracked from June through August 2006 among three lakes of varying flooding frequency, in the Mackenzie Delta. Microscopic image analysis showed spectrophotometric methods generally over-estimated TEP concentrations in this system. TEP concentrations were highest immediately after river flooding and declined through August. Contrary to expectation, TEP concentrations were highest in the high flood frequency lake (lowest dissolved organic carbon). TEP accounted for an average of ~83.7% of particulate organic carbon in the lakes during summer, 2006. Total suspended bacterial abundances ranged from ~10^6 to 10^7 cells/ml. Microscopic partitioning of this community estimated that free-living cells, TEP-attached, and other-attached bacteria respectively accounted for 13.9, 9.6, and 76.5% of total suspended bacteria. TEP-attached bacterial densities increased over the summer and were higher in low flood frequency lakes. Results suggest TEP-attached bacterial density is related to the abundance of favourable colonization sites.

Keywords: transparent exopolymer particles; TEP; Mackenzie Delta; attached bacteria; bacteria, aggregation

Subject Terms: limnology – Northwest Territories – Mackenzie Delta region; lake ecology – Northwest Territories – Mackenzie Delta region; Freshwater microbiology; Microbial populations; Water – Microbiology

ACKNOWLEDGEMENTS

While there is only one name on the title page of this thesis, there are many people who have contributed in a number of different ways. First, I must thank Lance Lesack for bringing me into his lab and giving me the opportunity to work in the Mackenzie Delta. As my senior supervisor, Lance has been very supportive. My skills as a scientist and limnologist have been well served under his guidance. I sincerely appreciate Lance allowing me the freedom to choose, develop, and complete a project that was truly my own. His excellent advice, and at times, much needed encouragement have been indispensable throughout my degree.

Committee members Max Bothwell and Leah Bendell-Young provided much needed input at critical times throughout my degree. Max must be thanked for trusting me with his fancy microscope, camera, and image analysis system, and for being flexible about the return date. Max has also helped by asking tough questions and displaying a genuine enthusiasm about my project. Leah's questions generally come from a different angle which has helped round out my thesis and she has also forced me to think about the "big picture" implications of my results. Thanks to the external examiner Jeff Curtis for helpful comments and advice.

In addition to being great company under the midnight sun as well as in the lab, Suzanne Tank and Jolie Gareis, have provided much needed advice and second opinions regarding methods, references, and the Delta. I'd like to thank the Aurora Research Institute in Inuvik. William Hurst and Sharon Katz helped our field work go smoothly by providing/fixing boats and equipment and answering questions. Many thanks to our field assistants Emily Hines and Leah Honka who worked long and hard during the summer of 2006. I'd like to thank Alex Fraser, and B-Jae Kelly for equipment needs at SFU.

Financial support for the project was generously provided by Lance Lesack's NSERC grant, science horizons youth internship program, northern science and training program, and the polar continental shelf project.

To the friends I've met in Vancouver, thank-you for giving me respite from the daily toils of a grad student. To the BISC department, thank-you for embracing a geography student as your own.

Thank-you to my family for always being there when I need you. Thank-you Mom and Dad for your unwavering love, support, and trust. Leslie and Steve, you guys provide me with an unlimited source of laughter and fun. To friends and family in Ontario, thanks for your sporadic e-mails or phone calls which make me feel like I'm still connected and in the loop.

Finally, Thank-you Heather, for accompanying me throughout the last 3 years. It's been amazing to have someone I trust close at all times. Your smile gives me plenty of light through rain, sleet, snow, and the occasional sunshine in Vancouver.

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1 GENERAL INTRODUCTION

1.1 Introduction

The Mackenzie River splits approximately 200 km from the Arctic Ocean, and forms a delta of intricately winding main and distributary channels. Within this extensive 13 000 km^2 delta, there are over 45 000 lakes causing significant changes to the river water discharged into the Beaufort Sea (Emmerton et al. 2007, Emmerton et al. 2008). Each spring, ice jamming results in extensive flooding delivering water, sediments, particulate organic matter (POM), dissolved organic matter (DOM), and nutrients to delta lakes (Mackay 1963). This input is extremely important as local precipitation is low resulting a negative water balance, through evaporation, for lakes that are not flooded (Bigras 1990).

A lake floods if water levels in the distributary channel exceed the lake-sill elevation, defined as the highest elevation along the connecting channel thalweg between the lake and the distributary channel (Marsh and Hey 1989). A lake classification suggested by Mackay (1963), includes three classes of lakes based on sill elevation. No-closure lakes are connected to the distributary channel throughout the open water season, low-closure lakes become detached from the distributary channel at some point during the open water season, and high-closure lakes do not flood every year. Marsh and Hey (1989) quantified these lakes and found no-, low-, and high-closure lakes represented 12, 55, and 33% of all Mackenzie Delta Lakes respectively. This classification has been adopted for the ecological study of the delta and will serve to separate lakes on the basis of flooding frequency with high, intermediate, and low flood-frequency lakes corresponding to no-, low-, and high-closure lakes respectively (Figure 1.1). Differences in frequency and duration of flooding drive chemical and biological gradients in lakes of differing sill elevation (Lesack et al. 1998). This ecosystem of naturally occurring gradients lends itself to hypothesis testing. In this study, we use this ecosystem to answer questions about a novel class of particles referred to as transparent exopolymer particles (TEP).

Transparent exopolymer particles (TEP) are ubiquitous in freshwater and marine ecosystems. TEP are discrete particles consisting of polysaccharide fibrils exuded by algae and bacteria. These otherwise invisible particles are visualized through staining

with the polysaccharide specific stain alcian blue (Alldredge et al. 1993). Their basic ecology has been studied in most types of marine systems. It is now known that TEP are important for organic matter flux (Passow et al. 2001, Engel et al. 2002), algal bloom termination (Passow et al. 1994, Logan et al. 1995), trace metal scavenging (Quigley et al. 2002), and transfer of carbon from the dissolved to particulate phase (Chin et al. 1998, Passow 2000) in some marine environments. Conversely, there is a relative dearth of information regarding the ecology of TEP in lacustrine ecosystems, especially when considering the diversity of lake ecosystems that exist. To date there have only been seven freshwater TEP studies (Table 1.1).

To our knowledge, TEP determination has not been performed in Mackenzie Delta Lakes or any other arctic, or floodplain lakes. To assess the potential importance of TEP in arctic floodplain lakes, we present the first data on the abundance and distribution of TEP in Mackenzie Delta Lakes. Our objectives were to determine the TEP concentrations and size distributions in lakes of differing flood frequency (sill elevation) and to assess whether patterns in TEP distribution were related to concurrently measured chemical and biological variables.

Microbial communities play an important role in Mackenzie Delta Lakes, potentially providing significant secondary production to support consumer communities. TEP are hotspots for microbial communities, and may be disproportionately important for the biogeochemical cycling of organic material. We will separate the bulk suspended bacterial community into free-living, TEP-attached, and other-attached bacterial components to assess which component harbours the greatest bacterial numbers. We will also discuss TEP-attached bacterial densities in the context of prevailing aquatic conditions.

1.2 Figures

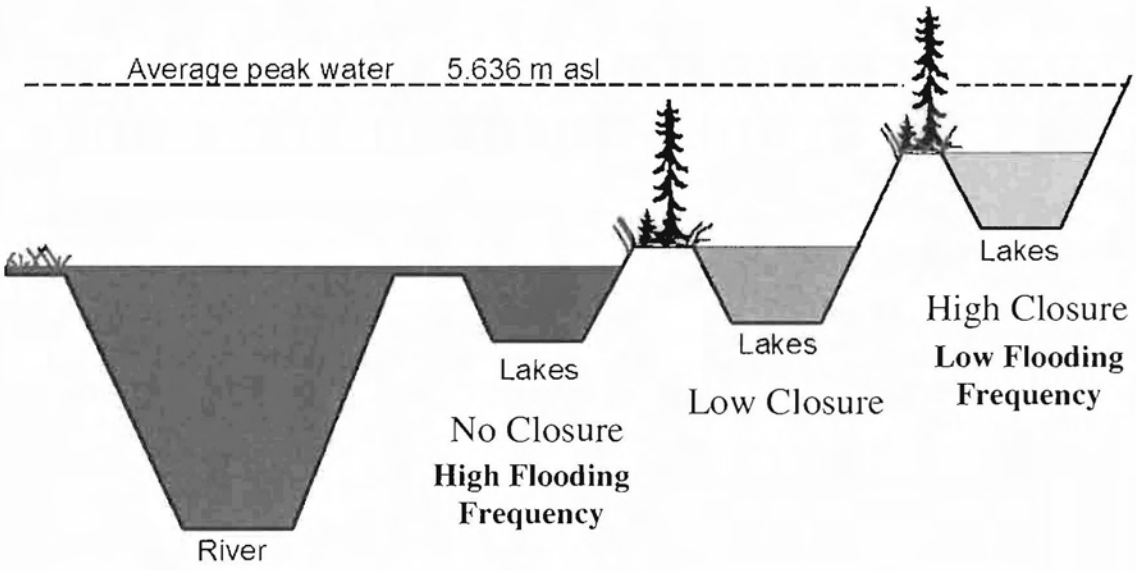


Figure 1.1 **Visual description of the lake classification system with an indication of the corresponding flooding frequency.**

1.3 Tables

Table 1.1 Summary of TEP research in lacustrine ecosystems.

Study Area	Title	Author
Lake Aydat and Pavin spring 2002	Community composition and activity of prokaryotes associated to detrital particles in two contrasting lake ecosystems.	(Lemarchand et al. 2006)
Lake Aydat and Pavin spring 2000	Distribution, size, and bacterial colonization of pico- and nano-detrital organic particles (DOP) in two lakes of different trophic status.	(Carrias et al. 2002)
Lake Constance summer/autumn 1996	Dynamics and bacterial colonization of microaggregates in a large mesotrophic lake.	(Brachvogel et al. 2001)
Lake Constance 1993	Formation of macroscopic organic aggregates (lake snow) in a large lake: The significance of transparent exopolymer particles, phytoplankton, and zooplankton.	(Grossart et al. 1997)
Lake Frederiksborg autumn 1994 summer 1995	Alcian blue-stained particles in a eutrophic lake.	(Worm and Sondergaard 1998a)
Lake Kinneret 1997-2000	Abundance and characteristics of polysaccharide and proteinaceous particles in Lake Kinneret.	(Berman and Viner-Mozzini 2001)
Lake Kinneret autumn (1995)	Occurrence and microbial dynamics of macroscopic organic aggregates (lake snow) in Lake Kinneret, Israel, in fall.	(Grossart et al. 1998)

1.4 References

Alldredge, A. L., U. Passow, and B. E. Logan. 1993. The abundance and significance of a class of large, transparent organic particles in the ocean. Deep-Sea Research Part I-Oceanographic Research Papers **40**:1131-1140.

Berman, T. and Y. Viner-Mozzini. 2001. Abundance and characteristics of polysaccharide and proteinaceous particles in Lake Kinneret. Aquatic Microbial Ecology **24**:255-264.

Bigras, S. C. 1990. Hydrological regime of lakes in the Mackenzie Delta, Northwest Territories, Canada. Arctic and Alpine Research **22**:163-174.

Brachvogel, T., B. Schweitzer, and M. Simon. 2001. Dynamics and bacterial colonization of microaggregates in a large mesotrophic lake. Aquatic Microbial Ecology **26**:23-35.

Carrias, J. F., J. P. Serre, T. Sime-Ngando, and C. Amblard. 2002. Distribution, size, and bacterial colonization of pico- and nano-detrital organic particles (DOP) in two lakes of different trophic status. Limnology and Oceanography **47**:1202-1209.

Chin, W. C., M. V. Orellana, and P. Verdugo. 1998. Spontaneous assembly of marine dissolved organic matter into polymer gels. Nature **391**:568-572.

Emmerton, C. A., L. F. W. Lesack, and P. Marsh. 2007. Lake abundance, potential water storage, and habitat distribution in the Mackenzie River Delta, western Canadian Arctic. Water Resources Research **43**.

Emmerton, C. A., L. F. W. Lesack, and W. F. Vincent. 2008. Mackenzie River nutrient delivery to the Arctic Ocean and effects of the Mackenzie Delta during open water conditions. Global Biogeochemical Cycles **22**.

Engel, A., M. Meyerhofer, and K. von Brockel. 2002. Chemical and biological composition of suspended particles and aggregates in the Baltic Sea in summer (1999). Estuarine Coastal and Shelf Science **55**:729-741.

Grossart, H. P., T. Berman, M. Simon, and K. Pohlmann. 1998. Occurrence and microbial dynamics of macroscopic organic aggregates (lake snow) in Lake Kinneret, Israel, in fall. Aquatic Microbial Ecology **14**:59-67.

Grossart, H. P., M. Simon, and B. E. Logan. 1997. Formation of macroscopic organic aggregates (lake snow) in a large lake: The significance of transparent exopolymer particles, phytoplankton, and zooplankton. Limnology and Oceanography **42**:1651-1659.

Lemarchand, C., L. Jardillier, J. F. Carrias, M. Richardot, D. Debroas, T. Sime-Ngando, and C. Amblard. 2006. Community composition and activity of prokaryotes associated to detrital particles in two contrasting lake ecosystems. Fems Microbiology Ecology **57**:442-451.

Lesack, L. F. W., P. Marsh, and R. E. Hecky. 1998. Spatial and temporal dynamics of major solute chemistry among Mackenzie Delta lakes. Limnology and Oceanography **43**:1530-1543.

Logan, B. E., U. Passow, A. L. Alldredge, H. P. Grossart, and M. Simon. 1995. Rapid formation and sedimentation of large aggregates is predictable from coagulation rates (half-lives) of transparent exopolymer particles (tep). Deep-Sea Research Part Ii-Topical Studies in Oceanography **42**:203-214.

Mackay, J. R. 1963. The Mackenzie Delta Area, NWT, Canada. Canadian Department of Mines and Technical Surveys, Geographical Branch **Memoir No. 8**:202pp.

Marsh, P. and M. Hey. 1989. The flooding hydrology of Mackenzie Delta Lakes near Inuvik, NWT, Canada. Arctic **42**:41-49.

Passow, U. 2000. Formation of transparent exopolymer particles, TEP, from dissolved precursor material. Marine Ecology-Progress Series **192**:1-11.

Passow, U., A. L. Alldredge, and B. E. Logan. 1994. The role of particulate carbohydrate exudates in the flocculation of diatom blooms. Deep-Sea Research Part I-Oceanographic Research Papers **41**:335-357.

Passow, U., R. F. Shipe, A. Murray, D. K. Pak, M. A. Brzezinski, and A. L. Alldredge. 2001. The origin of transparent exopolymer particles (TEP) and their role in the sedimentation of particulate matter. Continental Shelf Research **21**:327-346.

Quigley, M. S., P. H. Santschi, C. C. Hung, L. D. Guo, and B. D. Honeyman. 2002. Importance of acid polysaccharides for Th-234 complexation to marine organic matter. Limnology and Oceanography **47**:367-377.

Worm, J. and M. Sondergaard. 1998a. Alcian blue-stained particles in a eutrophic lake. Journal of Plankton Research **20**:179-186.

2 ASSESSING TRANSPARENT EXOPOLYMER PARTICLE (TEP) CONCENTRATION AMONG MACKENZIE DELTA LAKES USING TWO INDEPENDENT AND ACCEPTED METHODS

2.1 Abstract

During the ice-free season of 2006, we tested the spectrophotometric and microscopic methods as long-term sampling protocols for TEP determination in Mackenzie Delta Lakes. Both methods operate on the principle that TEP stain with alcian blue. The spectrophotometric method clearly overestimated TEP concentration with estimates ranging from 2830 to 27518 ± 9345 μg xanthan equiv/L corresponding to a mean 780% of the particulate organic carbon (POC) in the water column. The microscopic method yielded TEP estimates ranging from 4.8 to 295.8 ± 54.6 ppm and on average comprised ~84% of POC. After log transformation, TEP values obtained using the spectrophotometric method were significantly related ($r^2 = 0.58$, $p < 0.0001$, $df = 28$) to results obtained using the microscopic method. However, if this relationship is used to derive TEP estimates from the spectrophotometric method, the variance of these estimates are high. We consider the microscopic method for TEP determination in Mackenzie Delta Lakes the preferred method and caution researchers attempting to use the spectrophotometric method in ecosystems containing high concentrations of organic and inorganic material.

2.2 Introduction

Transparent Exopolymer particles (TEP) are ubiquitous in freshwater and marine ecosystems. The formation of TEP has been shown to be a significant pathway for the transfer of carbon from the dissolved to particulate phase. This previously unknown pathway has implications for the biogeochemical cycling of carbon (Passow and Alldredge 1995a, Passow 2002a). The bulk of the literature has focussed on TEP in marine systems and few measurements of TEP have been done in freshwater ecosystems.

The Mackenzie Delta is a novel, freshwater system in which to study TEP for a number of reasons. The Mackenzie River carries the highest sediment load of all rivers emptying into the Arctic Ocean (Milliman and Meade 1983). These sediment loads reach their greatest concentration during and immediately following spring break-up, characterised by extensive ice jamming (Marsh et al. 1999). The flood conditions associated with ice jamming result in sediment-laden and DOC-rich waters entering lakes, depending on lake-sill elevation (Mackay 1963). Although TEP analysis has been done on oceanic sediment traps, the material suspended in Mackenzie River floodwaters are likely to be very different than oceanic sediment trap material. Thus, applying the existing TEP assessment methodology to this, and other freshwater systems with high organic and inorganic suspended content may be problematic.

The Mackenzie Delta is characterized by its numerous small lakes that exhibit differences in chemical and biological composition. These differences depend on the frequency and duration of flood events, along with individual lake morphometry and the presence or absence of thermokarst activity. River water entering lakes during the spring flood re-initializes the lakes to varying degrees; some lakes are totally flushed while others only partially. River water delivers nutrients, sediments, POC, DOC to delta lakes and is therefore an important feature of delta lake dynamics. Subsequent changes that occur within a lake are then a product of the continuation or discontinuation of river water input, plus within lake processes.

There are currently two general methods by which TEP abundance are assayed. Both methods operationally define TEP as staining with alcian blue (pH ~2.5), which

selectively stains sulphated and carboxylated polysaccharide groups (Horobin 1988). A spectrophotometric method (Passow and Alldredge 1995b) measures the staining capacity of TEP compared to a TEP proxy. A microscopic method (Alldredge et al. 1993, Passow and Alldredge 1994) measures the abundance and size distribution of TEP after the particles have become visible from staining. The advantage of the spectrophotometric method is the speed at which samples can be analysed while the more laborious microscopic method yields more information (size distribution). In this paper, we will determine TEP abundance using the spectrophotometric method and verify our results using the microscopic method. We will make recommendations on the viability of both methods as long-term analysis protocols for TEP study in Mackenzie Delta Lakes.

2.3 Methods

2.3.1 Study Area

The Mackenzie watershed encompasses a huge area (1,805,200 km^2) including northern British Columbia, Alberta, and Saskatchewan, ranging north to the Beaufort Sea. Approximately 200 km from the Beaufort Sea, the Mackenzie river splits and forms a delta of intricately winding main and distributary channels. Within this extensive ~13 000 km^2 delta, there are over 45 000 lakes causing significant changes to river water chemistry before discharging to the Beaufort Sea (Emmerton et al. 2007, Emmerton et al. 2008).

Weekly samples were analysed from a set of 3 lakes located near the town of Inuvik, NT (68°19' N, 133°29'W). Lake 129 is a no-closure lake, Lake 56 is a high-closure lake and Lake 520 is a high closure lake with thermokarst activity. Point samples were analysed from another three lakes in the same region. Lake 80 is a no-closure lake, Lake 87 is a low-closure lake, and Lake 280 is a low-closure lake (Figure 2.1).

2.3.2 Sample Collection

During the summer of 2006 (early June – late August) whole water samples were obtained with a PVC tube sampler integrated over 1.5m from the lake surface (average lake depth <2m). Weekly samples were taken from the same location in each lake. Samples were stored in 1L HDPE bottles, and placed on ice in a cooler. Slides for microscopic analysis were prepared immediately upon return to the lab. Samples were filtered for spectrophotometric analyses immediately following slide preparation.

2.3.3 TEP Assessment

Relatively few aquatic ecologists have determined TEP concentrations in freshwater ecosystems. Although these determinations were apparently successful, we were apprehensive about how the methodology would perform in a floodplain ecosystem such as the Mackenzie Delta. Therefore, we used two accepted methods to determine TEP abundance and compared how the spectrophotometric method performed relative to

the microscopic method. The spectrophotometric assay described by Passow and Alldredge (1995b) is reported to be the most accurate method for TEP mass (Passow 2002a) but uses a TEP proxy to yield TEP mass estimates. The microscopic method (Alldredge et al. 1993) directly measures TEP (surface area), and yields more information because the size distribution of TEP is used in the determination.

2.3.3.1 Microscopic Method

Slide Preparation

Semi-permanent slides were prepared in duplicate based on the method of Alldredge et al (1993) with few modifications. 1-4ml samples were filtered through 25mm diameter, 0.4μm polycarbonate membrane filters at a constant filter pressure of no more than 150mm Hg. Samples were stained with 0.5ml of 0.2μm filtered alcian blue. The 0.03% alcian blue in 0.06% acetic acid was drawn through the filter immediately. The filter-transfer-freeze method (Hewes and Holm-Hansen 1983) was used to transfer filtered material to a glass slide. A loop of gel (0.035 g/ml gelatine, 25% glycerine in distilled water) was placed over the filtered material while still frozen and left to solidify. Prepared slides were stored at -20°C in sealed bags.

Assessment of Optimal Magnification for TEP Enumeration

Assessing TEP abundance via the microscopic method is a labour intensive endeavour requiring a significant amount of time to visually distinguish TEP particles from non-TEP particles in the sample. This is especially true for high sediment and particulate conditions that often occur in lakes of the Mackenzie Delta. It is generally accepted that the microscopic method systematically underestimates the TEP abundance due to difficulties associated with counting and sizing small TEP particles (Passow 2002a). The literature contains two basic approaches to TEP enumeration: sizing TEP manually under the microscope at a single magnification (Alldredge et al. 1993) or taking pictures at increasing magnifications and sizing TEP semi-automatically using an image analysis program (Mari and Kiorboe 1996). The latter method can be considered more robust because at lower magnifications, small TEP particles are difficult to see and size, whereas at high magnifications the larger, rare particles may be overlooked. Sizing TEP

particles at successive magnifications gives the best resolution, but is very time consuming. With these factors in mind, we decided to assess TEP abundance at multiple magnifications in order to determine which magnification gives the highest and therefore most accurate TEP abundance. For this comparison, TEP were enumerated at 100x, 250x, 400x, and 630x magnifications according to the protocol below.

Considering the theory that the microscopic method systematically underestimates TEP abundance, it follows that the magnification that gives the highest estimate of TEP abundance has minimized the errors associated with poor resolution of small TEP particles and the probability of missing large, rare particles. We found that TEP enumeration at 250x magnification provided the highest, and therefore best, estimate of TEP (Figure 2.2). Here we present TEP abundance as surface area/volume to avoid any bias introduced by converting surface areas to a volume concentration.

TEP Enumeration and Calculation of Size Distributions

A Moticam™ 1300 colour camera connected to a computer was used to capture digital images of 10 fields per slide at 250x magnification on a Leitz, Aristoplan microscope. Using Motic Images Advanced 3.0™ image analysis software, individual TEP were manually delineated to obtain the cross sectional area of TEP particles. From the cross sectional area of TEP particles, their equivalent spherical diameter (ESD) and volume (ESV) were calculated in order to place them into size categories and to calculate a volume concentration and size distribution.

The size distribution of TEP is described by the power law

$$\frac{dN}{d(d_p)} = k(d_p)^b$$

Where dN is the number of particles in size interval $d(d_p)$ (interval described by mean maximal ESD) and k and b are constants. b describes the shape of the size distribution such that as b decreases, the fraction of small particles decreases.

2.3.3.2 Spectrophotometric Method

The spectrophotometric method for TEP assessment was developed by Passow and Alldredge (1995b). It is a semi-quantitative method based upon the staining capacity of TEP by the polysaccharide specific stain alcian blue relative to the staining capacity per weight of a TEP proxy; in this case gum xanthan. The variable purity and solubility of alcian blue requires that all samples must be analysed using the same batch of dye. New dye solutions must be calibrated before use.

The gum xanthan standard was prepared by mixing 15 mg in 200 ml Nanopure™ water followed by grinding (10 strokes in a tissue grinder). The resulting solution is mixed for ~30 minutes before grinding again. This final solution should sit for another 15 minutes with occasional mixing. It is imperative that these steps are followed to ensure that the gel particles are fully hydrated before samples are filtered. The calibration curve will not be linear if the particles are not fully hydrated. Six replicate aliquots of 0.5, 1, 1.5, 2, and 3 ml of the standard solution were filtered through pre-weighed 47mm, 0.4μm pore size, polycarbonate membrane filters at a constant pressure of 150 mm Hg. These filters were dried overnight and reweighed on a microbalance (Cahn Electrobalance). Another six replicate aliquots of 0.5-3 ml of the standard solution were filtered as above before being stained with a 0.2 μm filtered solution of 0.02% alcian blue in 0.06% acetic acid. The stain was drawn through the filter immediately. Blanks were prepared as above using Nanopure™ water. Stained samples were dissolved in 80% sulphuric acid for at least 2 hours. Dissolving samples were gently mixed every half hour to eliminate air bubbles which can interfere with absorbance readings. Sample and blank absorbencies were measured at 787 nm on a spectrophotometer (Spectronic, Genesys 5).

The calibration factor, f_x is the inverse of the slope of the linear curve found by plotting gum xanthan weights (μg) vs. the corresponding absorptions (E_{787} - C_{787}) where E_{787} is the sample absorbance and C_{787} is the blank absorbance. The following equation is used to find the concentration of TEP (C_{TEP}) in μg gum xanthan equivalents per liter:

$$C_{TEP} = (E_{787} - C_{787}) \bullet (V_f)^{-1} \bullet f_x$$

where V_f is the volume filtered in litres, and f_x is the calibration factor in μg.

Lake water samples were filtered, stained, dissolved in sulphuric acid and read in a spectrophotometer as described above. The only change in protocol was that sample filters were sealed in aluminium foil and stored at -20°C for up to two weeks before dissolution. Filtered volumes ranged from 5-20 ml.

2.3.4 Calculating TEP Carbon Content

The amount of TEP carbon (TEP_{carbon}) present in a sample (ugC/L) was estimated using the empirically derived equation from Engel and Passow (2001),

$$TEP_{carbon} = 0.75 \bullet TEP_{colour}$$

where TEP_{colour} is the spectrophotometric estimate of TEP abundance in µg xanthan equiv/L.

To find TEP_{carbon} using the microscopic method, TEP carbon per particle (TEP_{pc}) is estimated using the empirical equation developed by Mari (1999)

$$TEP_{pc} = 0.25 \bullet 10^{-6} R^{2.55}$$

where TEP_{pc} is in µg and R is the equivalent spherical radius (µm). An estimate of the total amount of TEP carbon is found according to

$$TEP_{carbon} = \sum_{p=1}^{n} \left[k(d_p)^b \right] \bullet \left[0.25 \bullet 10^{-6} \bullet \left(\frac{d_p}{2} \right)^{2.55} \right]$$

where *k* and b are constants from the size distribution of TEP particles, d is the mean diameter (µm) of size class p and n is the number of size classes.

2.3.5 Particulate Organic Carbon and Total Suspended Solids

Particulate organic carbon (POC) was estimated using a loss on ignition method, so that the percent contribution of TEP to POC could be assessed. Lake water was filtered through a pre-weighed, pre-combusted Whatman GF/C, before being oven dried and stored in a desiccator until it was weighed to find the total suspended solids (TSS). The filter was then combusted at 550°C for 9 hours before being weighed again. The weight

difference between TSS and the combusted filter gives an estimate of particulate organic matter (POM). The POM (mg) estimate is multiplied by a factor of 0.47 ± 0.01 (Dean 1974) to obtain an estimate of POC in the volume filtered (mg/L).

2.4 Results

2.4.1 Spectrophotometric Method

The spectrophotometric method, developed by Passow and Alldredge (1995b), yielded extremely high estimates of TEP abundance, ranging from 2830 to 27518 ± 9345 μg xanthan equiv/L, in the lakes surveyed. The method estimates that on average 769% of the POC is in the form of TEP. This indicates that there are errors associated with the determination of TEP concentration and/or TEP_{carbon} using the spectrophotometric method in Mackenzie Delta Lakes. Engel and Passow (2001) found good correlation between TEP_{carbon} derived from microscopic determinations and spectrophotometrically estimated TEP ($r^2 = 0.80$, n = 90). The relationship we found, for log transformed data (Figure 2.3B), was also significant but there was more variability as indicated from the lower coefficient of determination ($r^2 = 0.62$, n = 29). One value was excluded from the analysis as it was an obvious outlier.

2.4.2 Microscopic Method

After converting to a volume concentration, TEP abundance ranged from 4.8 to 295.8 ± 54.6 ppm. TEP_{carbon} was estimated as ranging from 171 – 3242 ± 677 μg C/L and on average accounted for ~84% of POC.

2.4.3 Method Comparison

The general trends in TEP abundance observed using both methods are very similar (Figure 2.4). TEP abundance is high immediately after flooding and then drops quickly and becomes relatively stable throughout the remainder of the sampling period. However, there is an inconsistent difference in relative TEP abundance estimated using the two methods (Figure 2.4).

It is apparent that the spectrophotometric method overestimates TEP in this type of freshwater system. However, the microscopic method is too laborious to adopt as a long-term sampling protocol. We attempted to calibrate the spectrophotometric method by plotting observed TEP abundances (μg xanthan equiv/L) as a function of TEP volume

concentration (ppm), measured using the microscopic method (Figure 2.3). If the correlation is strong, the spectrophotometric method may be used by applying its results to the calibration curve.

After log transforming both spectrophotometric and microscopic TEP estimates, a significant relationship is found ($r^2 = 0.58$, $p < 0.0001$, $df = 28$) (Figure 2.3A). The relationship is improved marginally ($r^2 = 0.62$, $p < 0.0001$, $df = 28$) if microscopic TEP estimates are converted to TEP_{carbon} estimates (Figure 2.3B). It appears that, at best, the calibration curve could only be useful for identifying broad trends in TEP abundance.

2.5 Discussion

The only precedence for TEP abundance values of the magnitude estimated here, by the spectrophotometric method, come from the massive gel phenomenon documented periodically in the Northern Adriatic Sea (Passow 2002a). While TEP concentrations of this magnitude are evidently possible under some conditions, it is not possible for TEP concentrations to account for ~770% of total suspended POC. It is expected that some smaller TEP particles will pass through filters used to assess POC (Whatman GF/C), but it is unlikely that these particles could account for such a large overestimation. Engel and Passow's (2001), empirically derived relationship between TEP_{colour} and TEP_{carbon} may not be applicable in Mackenzie Delta Lakes, as it was developed using a natural oceanic population, under lab conditions. This could contribute error to our TEP carbon values, but it is unlikely that it would account for the large discrepancies found here. Filter clogging (0.2 μm, polycarbonate) could account for the extremely high TEP abundance estimates obtained with the spectrophotometric method, however, this is unlikely, as relatively small volumes were filtered through 47 mm diameter filters.

TEP volume concentrations estimated using the microscopic method were well within the range reported for both marine and freshwater ecosystems (See Passow 2002a). Since TEP appeared to account for an average of ~84% of POC, the formation of TEP could be a significant pathway for DOC transfer to POC in Mackenzie Delta Lakes. However, our estimates of TEP_{carbon} are not without uncertainty. TEP_{carbon} was estimated using an equation derived from bubbling lab cultures, under varying conditions, for a single oceanic phytoplankton species, *Thalassiosira weissflogii*. The issue here is whether or not 'natural' TEP have the same fractal dimension as the 'artificial' TEP used to derive the equation. We concede that the equation may not be totally applicable to our data, as we have measured TEP in freshwater, that presumably formed through an alternate mechanism (coagulation), involving precursors from a natural and mixed algal community, under different environmental conditions than those used to derive the equation. However, fractal dimensions similar to that used in the TEP_{carbon} equation here have been observed for 'natural' TEP (Mari and Burd 1998). Although the fractal dimension of TEP has also been estimated to be as low as 1.5 (Mari and Burd 1998), this

estimate was based on a non conservative tracer (bacterial colonization of different size classes of TEP). In the absence of data allowing the direct calculation of TEP_{carbon} for this study, the equation is useful for estimating the proportion of carbon that could be present as TEP in the Mackenzie Delta, although our apparent TEP_{carbon} values are likely overestimates.

The reasons for such a large over estimation of TEP using the spectrophotometric method are not totally clear. The highest TEP values obtained came from sediment laden waters directly after the flood. This result is unexpected, as Passow et al (2001) found that lithogenic material found in their sediment traps covered TEP, and physically blocked alcian blue from binding to polysaccharides. This suggests that the large amount of lithogenic material suspended in flood waters should have caused an underestimation of TEP, which was clearly not the case. As stated earlier, suspended material in Mackenzie Delta waters is likely very different than the material collected in oceanic sediment traps thus the lithogenic material found in the Mackenzie Delta may not be comparable to that found in oceanic sediment traps.

The chemical properties of alcian blue include an extremely low diffusivity due to its high molecular weight (Horobin 1988). This means that although polysaccharide binding sites on the exterior of TEP particles will be instantly stained with alcian blue, polysaccharides within TEP may not bind alcian blue, depending on TEP porosity. Thus there is uncertainty associated with the penetration of the stain compared to the gel proxy. The time required for complete staining, defined as alcian blue binding to all sulphated and carboxylated polysaccharide sites within the particle, will be related to the size distribution and porosity of TEP, which is impossible to control for. TEP is hydrated (~99% water) and generally believed to be quite porous so stain penetration time is likely to be fast. Longer staining times would ensure complete stain penetration but are not advisable as phytoplankton cells lyse after 5 seconds in the staining solution, potentially releasing stainable material (Passow and Alldredge 1995b). Incomplete staining is unlikely, considering the swollen nature of TEP particles, but differences between naturally occurring TEP and the gel proxy, xanthan gum, used to calibrate the spectrophotometric method may introduce errors into the analysis.

If alcian blue stains non-TEP suspended material, the spectrophotometric method would over estimate actual TEP abundances. It was evident from the microscopic analysis, that non-TEP particles (mainly detritus) were sometimes stained along with TEP particles. Stained detrital particles were not included in microscopic counts, as they were distinguished from TEP, but could not be eliminated from the spectrophotometric determinations. In some cases phytoplankton cell mucilage was also stained, adding to the overestimation of the spectrophotometric method, but again, would not be included in the microscopic counts. This is a likely source of much of the overestimation associated with the spectrophotometric method, although we cannot quantify the error. The high value of the y-intercept (Figure 2.3) in the relationship between the spectrophotometric method and the microscopic method gives an indication that TEP estimated by the spectrophotometric method is inflated by non-TEP particles that stain with alcian blue. Also, the inconsistencies associated with the difference between the two methods, as shown in Figure 2.4, may represent a measure of changes in alcian blue stainable, non-TEP material.

Within the emerging field of aquatic gel particles, there has been substantial variability associated with particle definition. Gel particles are generally defined operationally by one or more defining properties, such as staining with a compound specific stain (transparent exopolymer particles, alcian-blue particles, DAPI yellow particles, coomassie stained particles), or based on physico-chemical properties (self-assembled microgels, self-assembled micelle-like microparticles) (Alldredge et al. 1993, Long and Azam 1996, Chin et al. 1998, Worm and Sondergaard 1998a, Carrias et al. 2002, Kerner et al. 2003). While operational definitions work well within respective classifications, there is little information regarding the degree of overlap between different operationally defined particle types. It would be naïve to believe that particles within each classification are so distinct that they would be totally excluded from other gel particle categories. With this in mind, different operational definitions, and the methods associated with them, can provide certain advantages which make them more or less suitable for certain types of research and research goals. The broadly defined TEP have been reliably used to assess the implications of abiotically and biotically assembled gel particles for aggregation of diatom blooms, vertical transport of POC, and potential

for microbial interactions with gel particles (Passow and Alldredge 1994, 1995a, Azetsu-Scott and Passow 2004). The more rigorously defined self-assembled microgels have specific fluorescence properties allowing them to be separated using a modified flow-cytometry system (Orellana et al. 2007). This will prove to be a powerful tool in advancing our knowledge of the chemical composition of gel particles, and help elucidate the relationship between DOC and POC, as well as the persistence of refractory DOC in aquatic ecosystems.

2.6 Conclusion

The main advantage of the spectrophotometric method for estimating TEP abundance is its ease of implementation, quick analysis times, and high sample throughput. Conversely, the main drawbacks of the microscopic method for TEP abundance is the time consuming nature of slide preparation and image analysis. The microscopic method requires the estimation of TEP size distribution which is important for estimates of TEP-attached bacterial abundances, TEP formation mechanisms, and particle dynamics. Therefore the microscopic method yields important information, especially when research goals include ecological questions. A researcher's choice of TEP abundance estimation method will depend on the research question, the accuracy needed, the system in which they are working, and potential time constraints on analysis.

It is our opinion that without specific modifications, the spectrophotometric method should not be used for TEP determination in Mackenzie Delta Lakes due to vast overestimates compared to the microscopic method. A calibration curve yielded a loose relationship between spectrophotometrically and microscopically determined TEP. However, due to the variability associated with the relationship, the curve cannot be used reliably for TEP determination beyond rough estimation.

2.7 Figures

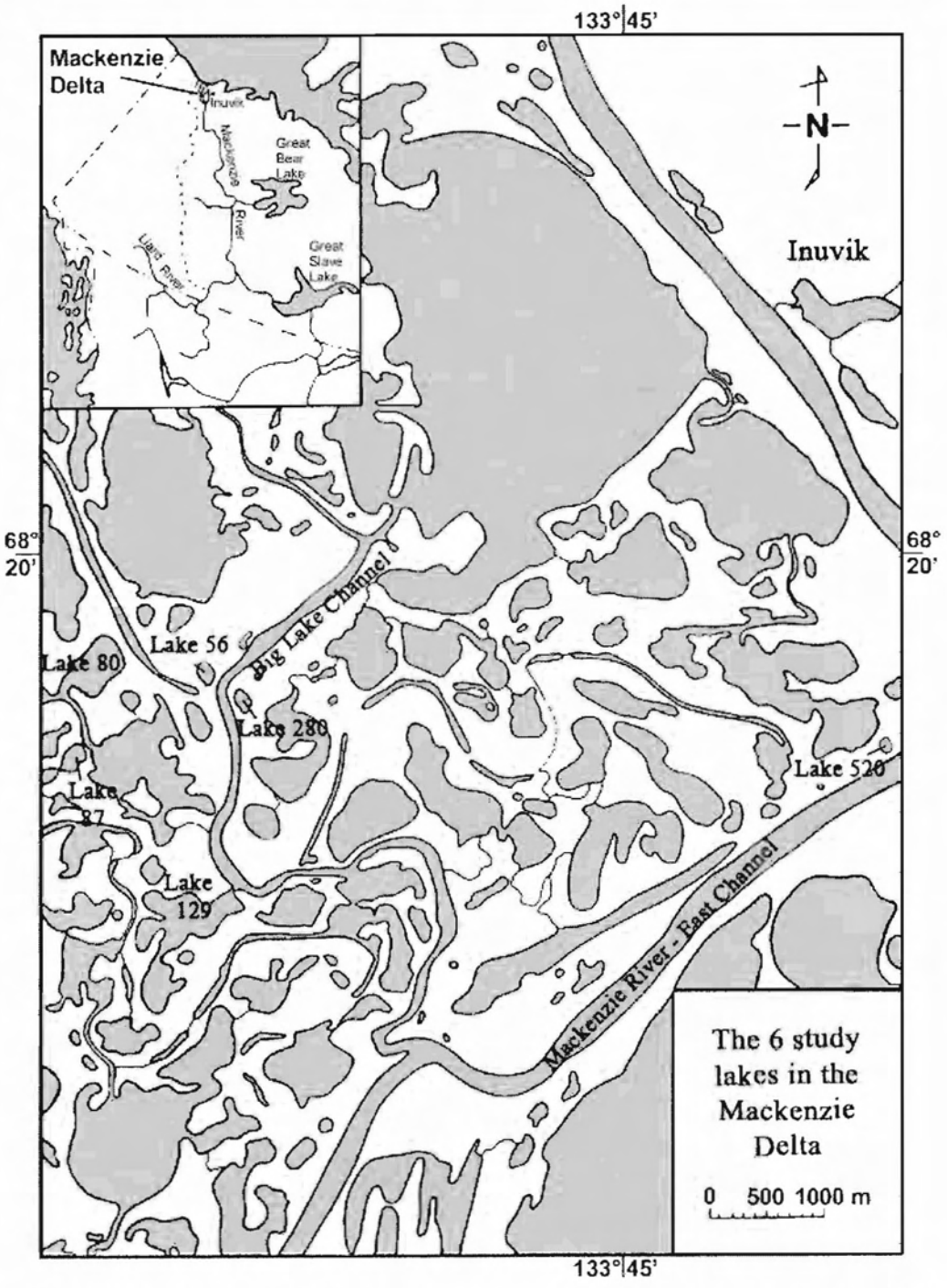


Figure 2.1 The six lakes included in the study are shown in a subsection of the Mackenzie Delta located near the town of Inuvik.

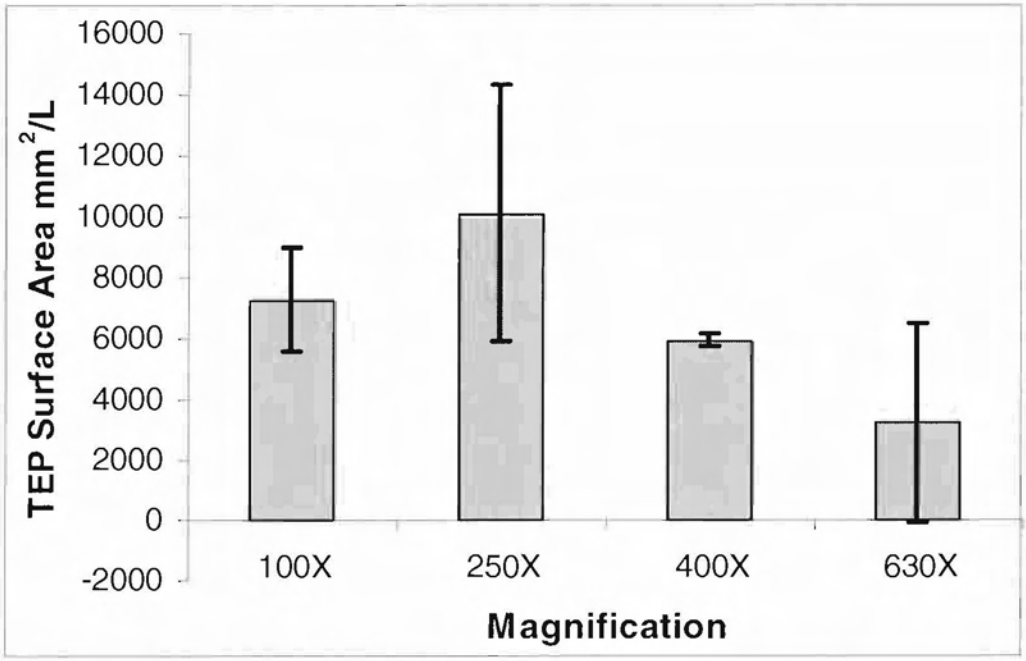


Figure 2.2 **Comparison of total TEP surface area at the four magnifications tested to determine the single magnification that optimizes TEP determination. Error bars are 95% confidence intervals.**

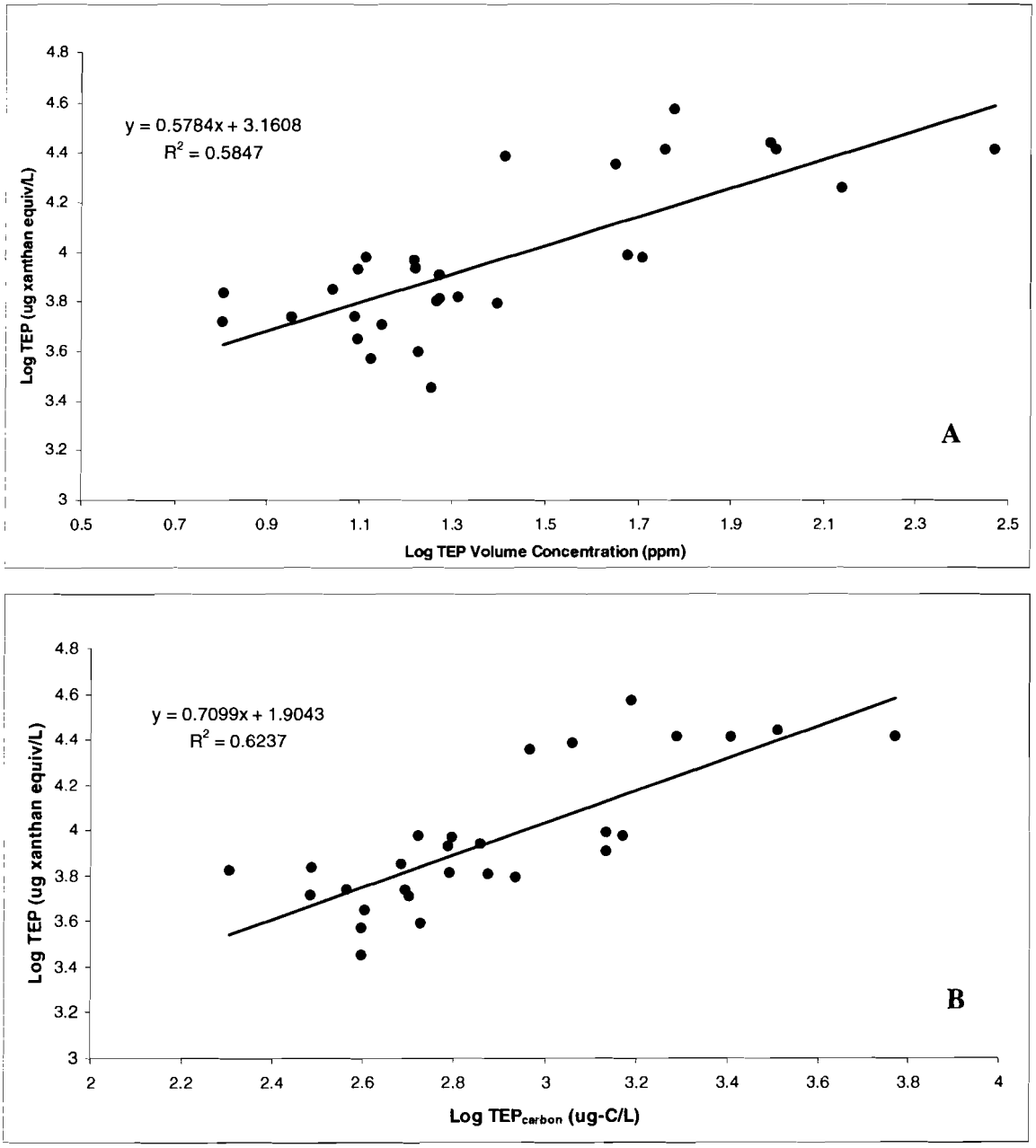


Figure 2.3 **Spectrophotometrically measured TEP (µg xanthan equiv/L) is plotted against (A) TEP volume concentration (ppm) and (B) TEP_{carbon} (µg-C/L). All data are Log transformed.**

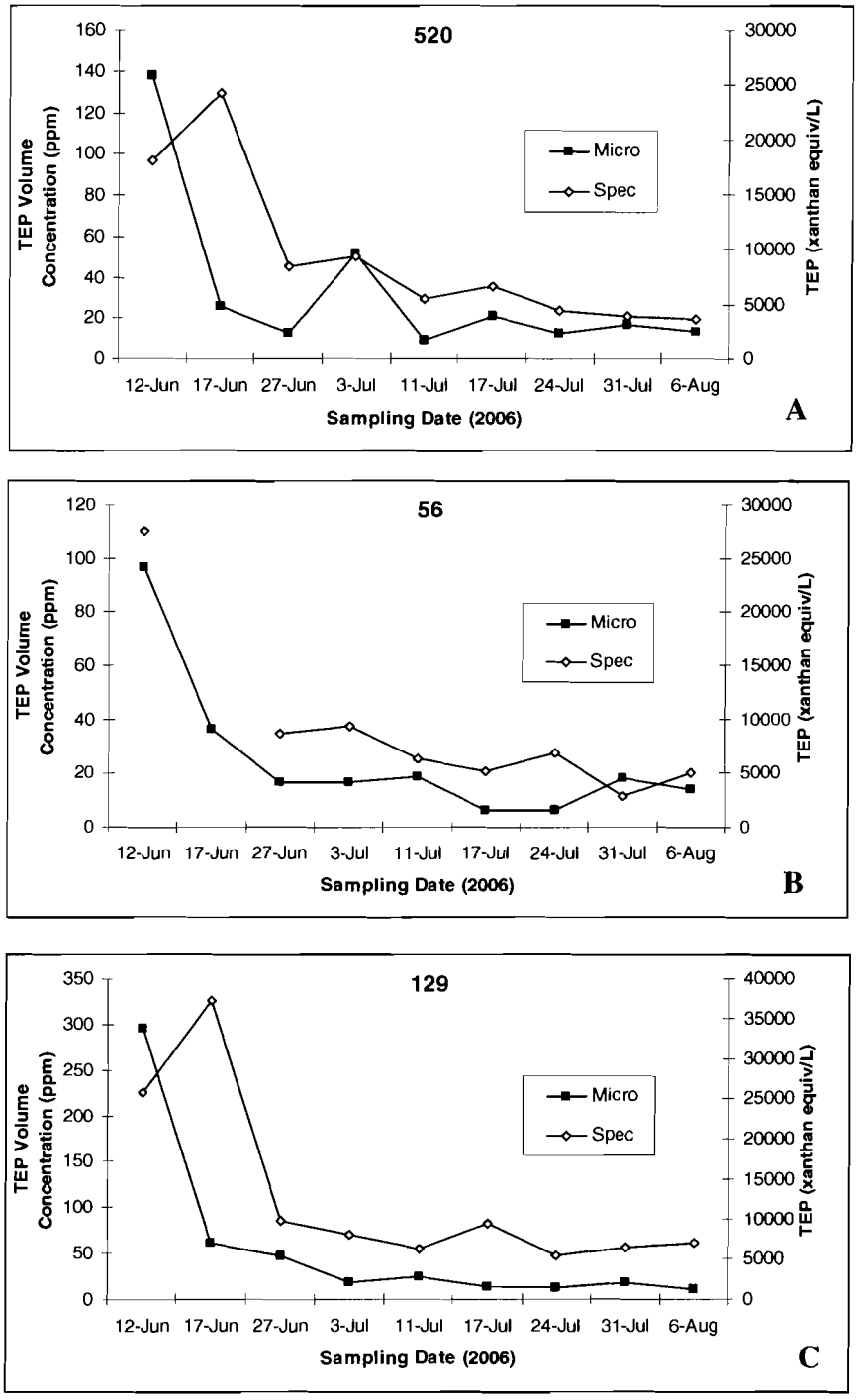


Figure 2.4 **Seasonal abundance of TEP as measured by the spectrophotometric method (µg xanthan equiv/L) and the microscopic method (ppm) in lakes A) 520, B) 56, and C) 129 during the open water season of 2006.**

2.8 References

Alldredge, A. L., U. Passow, and B. E. Logan. 1993. The abundance and significance of a class of large, transparent organic particles in the ocean. Deep-Sea Research Part I-Oceanographic Research Papers **40**:1131-1140.

Azetsu-Scott, K. and U. Passow. 2004. Ascending marine particles: Significance of transparent exopolymer particles (TEP) in the upper ocean. Limnology and Oceanography **49**:741-748.

Carrias, J. F., J. P. Serre, T. Sime-Ngando, and C. Amblard. 2002. Distribution, size, and bacterial colonization of pico- and nano-detrital organic particles (DOP) in two lakes of different trophic status. Limnology and Oceanography **47**:1202-1209.

Chin, W. C., M. V. Orellana, and P. Verdugo. 1998. Spontaneous assembly of marine dissolved organic matter into polymer gels. Nature **391**:568-572.

Dean, W. E. 1974. Determination of carbonate and organic-matter in calcareous sediments and sedimentary-rocks by loss on ignition - Comparison with other methods. Journal of Sedimentary Petrology **44**:242-248.

Emmerton, C. A., L. F. W. Lesack, and P. Marsh. 2007. Lake abundance, potential water storage, and habitat distribution in the Mackenzie River Delta, western Canadian Arctic. Water Resources Research **43**.

Emmerton, C. A., L. F. W. Lesack, and W. F. Vincent. 2008. Mackenzie River nutrient delivery to the Arctic Ocean and effects of the Mackenzie Delta during open water conditions. Global Biogeochemical Cycles **22**.

Engel, A. and U. Passow. 2001. Carbon and nitrogen content of transparent exopolymer particles (TEP) in relation to their alcian blue adsorption. Marine Ecology-Progress Series **219**:1-10.

Hewes, C. D. and O. Holm-Hansen. 1983. A method for recovering nanoplankton from filters for identification with the microscope - The filter-transfer-freeze (ftf) technique. Limnology and Oceanography **28**:389-394.

Horobin, R. W. 1988. Understanding histochemistry. Selection, evaluation and design of biological stains. Wiley.

Kerner, M., H. Hohenberg, S. Ertl, M. Reckermann, and A. Spitzy. 2003. Self-organization of dissolved organic matter to micelle-like microparticles in river water. Nature **422**:150-154.

Long, R. A. and F. Azam. 1996. Abundant protein-containing particles in the sea. Aquatic Microbial Ecology **10**:213-221.

Mackay, J. R. 1963. The Mackenzie Delta Area, NWT, Canada. Canadian Department of Mines and Technical Surveys, Geographical Branch **Memoir No. 8**:202pp.

Mari, X. and A. Burd. 1998. Seasonal size spectra of transparent exopolymeric particles (TEP) in a coastal sea and comparison with those predicted using coagulation theory. Marine Ecology-Progress Series **163**:63-76.

Mari, X. and T. Kiorboe. 1996. Abundance, size distribution and bacterial colonization of transparent exopolymeric particles (TEP) during spring in the Kattegat. Journal of Plankton Research **18**:969-986.

Marsh, P., L. F. W. Lesack, and A. Roberts. 1999. Lake sedimentation in the Mackenzie Delta, NWT. Hydrological Processes **13**:2519-+.

Milliman, J. D. and R. H. Meade. 1983. World-wide delivery of river sediment to the oceans. Journal of Geology **91**:1-21.

Orellana, M. V., T. W. Petersen, A. H. Diercks, S. Donohoe, P. Verdugo, and G. van den Engh. 2007. Marine microgels: Optical and proteornic fingerprints. Marine Chemistry **105**:229-239.

Passow, U. 2002a. Transparent exopolymer particles (TEP) in aquatic environments. Progress in Oceanography **55**:287-333.

Passow, U. and A. L. Alldredge. 1994. Distribution, size and bacterial-colonization of transparent exopolymer particles (tep) in the ocean. Marine Ecology-Progress Series **113**:185-198.

Passow, U. and A. L. Alldredge. 1995a. Aggregation of a diatom bloom in a mesocosm - The role of transparent exopolymer particles (tep). Deep-Sea Research Part Ii-Topical Studies in Oceanography **42**:99-109.

Passow, U. and A. L. Alldredge. 1995b. A dye-binding assay for the spectrophotometric measurement of transparent exopolymer particles (TEP). Limnology and Oceanography **40**:1326-1335.

Passow, U., R. F. Shipe, A. Murray, D. K. Pak, M. A. Brzezinski, and A. L. Alldredge. 2001. The origin of transparent exopolymer particles (TEP) and their role in the sedimentation of particulate matter. Continental Shelf Research **21**:327-346.

Worm, J. and M. Sondergaard. 1998a. Alcian blue-stained particles in a eutrophic lake. Journal of Plankton Research **20**:179-186.

3 TEP CONCENTRATION AND SIZE DISTRIBUTION AMONG MACKENZIE DELTA LAKES

3.1 Abstract

We assessed transparent exopolymer particle (TEP) concentrations in 3 lakes of differing flooding frequency from June through August in 2006 and 2007. A seasonal pattern of high post-flood TEP concentration and low late summer TEP concentration was present in both years. However, during the low flood year (2007), post-flood TEP abundances were an order of magnitude lower than during the high flood year (2006), suggesting that flood magnitude mediates post-flood TEP concentration. We hypothesised that TEP concentration would increase with decreasing flooding frequency due to typical chemical and biological gradients. Contrary to this hypothesis, there was no statistical difference in TEP concentration between lakes of differing flooding frequency, though mean concentrations appeared to be highest in a high flood frequency lake. TEP contributed an average of 84% of POC during 2006 and likely represents a significant pathway for carbon cycling in Mackenzie Delta Lakes. The spectral slope of TEP size distributions showed that small TEP particles contributed a greater proportion of TEP volume concentration on separate occasions in separate lakes. This resulted in rapid aggregation and return to a small particle depleted state on both occasions. Regression of relevant variables showed the importance of river water inputs, DOC, and bacteria for TEP concentrations in Mackenzie Delta Lakes. The strong correlation between TEP and CDOC indicates that some TEP may be formed from terrestrial sources.

3.2 Introduction

The identification of new particle classes, including transparent exopolymer particles (Alldredge et al. 1993), Coomassie blue particles (Long and Azam 1996), and DAPI yellow particles (Mostajir et al. 1995) in marine and freshwater ecosystems has led to numerous successful studies, contributing to our burgeoning knowledge of the ecosystems in which they are found. Transparent exopolymer particles (TEP) are the most widely studied class of gel particles and are ubiquitous in aquatic environments.

TEP are formed through aggregation of colloidal dissolved organic matter (DOM) to sub-micron particles and further aggregation to particulate organic matter (POM), or TEP, which can be isolated using polycarbonate filters (Passow 2002a). Specifically, polysaccharide fibrils form TEP precursors; complex networks which are stabilized by physical and chemical crosslinks as well as water itself. Indeed, TEP particles are hydrogels and are assumed to be ~99% water (Chin et al. 1998). Physical stabilization comes from the entanglement of fibrillar polysaccharides, and chemical stabilization from divalent cationic (Ca^{2+} and Mg^{2+}) bonds (Kloareg and Quatrano 1988) and hydrogen bonds (Chin et al. 1998). Through these mechanisms, TEP-precursors aggregate to sub-micron particles which continue to aggregate to form TEP. The distinction between TEP precursors, submicron particles, and TEP is important because TEP precursors and submicron particles behave as colloids, and TEP as particles (Leppard 1995).

According to Chin et al. (1998), approximately 10% of DOM will form spontaneously assembled particles. The DOM necessary for TEP formation is mucilaginous material, excreted by a variety of organisms, for a variety of species specific purposes (Decho 1990, Wotton 2005). The composition and amount of excretion can be variable depending on the species, as well as their physiological and environmental conditions (Leppard 1995, Myklestad 1995). Although the source of this material is biotic, the aggregation is abiotic, which changes the traditional view of bacteria mediating all transfer of DOM to POM. Phytoplankton have been identified as being particularly important for the release of these fibrillar exudates (Myklestad 1995). Laboratory based studies have found that bacteria also have the potential to release TEP

forming precursors (Stoderegger and Herndl 1999, Passow 2002a, Radic et al. 2003, Radic et al. 2006). However, in situ studies generally find little evidence for direct contributions by the bacterial community, and suggest bacteria are important for TEP production through exudate modification or TEP precursor scavenging (Grossart and Simon 2007, Sugimoto et al. 2007).

The Mackenzie Delta is characterized by numerous shallow lakes, in a stunted black spruce boreal to tundra ecosystem with underlying permafrost. Each spring, the Mackenzie River is jammed with ice creating flood conditions for lakes depending on the flood stage of the river as well as the sill elevation of the individual lake (Mackay 1963, Marsh and Hey 1989). This flood acts as recharge for flooded lakes by delivering water, sediments, nutrients, POC, and DOC. Differences in lake-sill elevation cause differences in flood frequency and duration, which in turn create gradients in chemical and biological variables. Low sill elevation lakes are flooded frequently and for a longer duration, while high sill lakes are flooded infrequently (not every year), and for a short duration. A set of lakes with different sill elevations is a convenient system in which to examine TEP variation with changing chemical and biological variables.

Variables of particular importance to TEP concentrations include TSS, DOC, carbohydrates, bacterial abundance, microalgal abundance and macrophyte abundance. We formulated hypotheses on how these variables would affect TEP and where TEP concentrations would likely be greatest among Mackenzie Delta Lakes. We hypothesised that TEP concentration would increase with:

1) decreasing total suspended solids (TSS), as suspended sediments would cause TEP concentration to decrease through aggregation and sedimentation;

2) increasing dissolved organic carbon (DOC) and dissolved carbohydrate concentrations, as TEP-precursors are thought to be a part of these dissolved fractions;

3) increasing microalgal abundance, as microalgae, and especially phytoplankton, have been shown to contribute TEP-precursors;

4) increasing macrophyte abundance, as macrophytes may contribute TEP precursors; and

5) increasing bacterial abundance as bacteria generally have a net positive effect on TEP concentrations.

Other factors that could affect TEP concentrations include wind mixing (turbulence) and ultraviolet radiation. Turbulence can cause increases in TEP concentrations via increasing coagulation rates, while UV has been shown to inhibit the formation of self assembled gels (Orellana and Verdugo 2003), although it is not clear whether this significantly affects TEP concentrations.

The variables above, generally, follow a trend among lakes that depends on the flood frequency and duration of the lakes, and thus, their sill elevation. Our hypotheses are summarized in Figure 3.1. In Mackenzie Delta Lakes, TSS increases with decreasing sill elevation (Marsh et al. 1999). DOC increases with increasing sill elevation (Squires and Lesack 2003a) and we hypothesise that carbohydrates will follow the trend for DOC, although carbohydrates have not been measured in the Mackenzie Delta thus far. Microalgal and macrophyte biomass also increase with increasing sill elevation (Squires et al. 2002a). Previous studies have not found a consistent pattern between bacterial abundances and sill elevations among Mackenzie Delta Lakes. Wind mixing and turbulence increases with decreasing sill elevation as lower elevation lakes tend to have a greater surface area. UV penetration increases with increasing sill elevation as higher elevation lakes are more clear (Gareis 2007). Due to study limitations we did not measure periphyton and macrophyte biomass, in situ turbulence, or UV penetration, however, we feel confident these variables follow the trends we have described.

Although we do not know the magnitude of the relationships between TEP and the variables above, all variables except wind mixing (turbulence) and UV penetration suggest TEP concentration will increase with increasing sill elevation. Therefore, we hypothesise that TEP concentrations will increase with increasing sill elevation (decreasing flood frequency) in Mackenzie Delta Lakes.

In the present study we test the above hypotheses and assess seasonal changes in TEP abundance over the ice-free period of the summer. We also assess whether TEP size distributions vary seasonally, or exhibit variation in lakes of different flood frequency and duration. These questions will add to our limited knowledge of TEP ecology in freshwater ecosystems and provide a basis for further study in an interesting and unique study location.

3.3 Methods

3.3.1 Study Area

The Mackenzie watershed encompasses a huge area including northern British Columbia, Alberta, and Saskatchewan, ranging north to the Beaufort Sea. Approximately 200 km from the Beaufort Sea, the Mackenzie River splits and forms a delta of intricately winding main and distributary channels. Within this extensive 13 000 km^2 delta, there are over 45 000 lakes causing significant changes to water chemistry (Emmerton et al. 2007, Emmerton et al. 2008) before being discharged into the Beaufort sea. The Mackenzie River and Delta remain frozen for over half the year. During the spring, melt-water from the southern portion of the basin travels to the northern reaches of the Mackenzie where it is met by ice. The result is ice jamming and extensive flooding within the delta (Mackay 1963). The flood waters of the Mackenzie River carry large amounts of sediment, and dissolved organic and inorganic matter which is deposited on the banks, levies, and in the lakes of the delta (Carson et al. 1999). Delta lakes are differentially flooded depending on lake sill elevation and river stage such that lakes will be flooded frequently, intermediately, and infrequently based on whether or not the lake is connected to the river for the duration of the summer (no-closure), becomes detached from the river during the summer (low-closure), or is not necessarily flooded every year (high-closure) (Mackay 1963, Marsh and Hey 1989, 1994). This initial flood drives many of the chemical and biological gradients found in the Mackenzie Delta along the flood frequency continuum. Lakes included in the present study were chosen based on accessibility, sill elevation, and knowledge available from previous research. Lake 129 is a frequently flooded, no closure (2.36 m asl) lake; 56 is an infrequently flooded, high-closure (4.62 m asl) lake, and 520 is an infrequently flooded, high-closure (4.91 m asl) lake with thermokarst activity. All lakes are located near the town of Inuvik, NT (68°19' N, 133°29'W).

3.3.2 Sample Collection

During the summer of 2006 and 2007 (early June – late August), whole water samples were obtained with a tube sampler, integrated over a depth of 1.5 m from the lake surface (average lake depth < 2 m). Samples were taken from the same location in

each lake at weekly intervals during 2006 and 4 times over the same time frame in 2007. It is assumed that Mackenzie Delta Lakes are well mixed during the open water season and these point samples were taken as representative values of the chemical and biological measurements made. Samples were stored in 1L HDPE bottles and placed on ice in a cooler. Samples were processed immediately upon return to the lab.

3.3.3 Chlorophyll a, TSS, POC, TDOC, CDOC

Lake water for chlorophyll a analysis was vacuum filtered at no more than 175 mm Hg through Whatman, GF/C filters. The filters were then frozen at -20°C until subsequent analysis. Extraction of Chl a was performed in hot 90% ethanol which was then refrigerated in the dark for 24 hours before reading on a Cary, Eclipse fluorometer at an excitation of 436 nm (slit 10 nm), emission of 680 nm (slit 5 nm) in a 1 cm path length, quartz cuvette. A calibration curved was produced using serial dilutions of Chl a (Sigma, C5753) in 90% ethanol.

Lake water was filtered through combusted, pre-weighed GF/C filters and TSS (mg/L) was determined gravimetrically. These filters were then combusted at 550°C for 9 hours (time required to reach stable weight) before being weighed again. The filter's combusted weight was subtracted from its dry weight to obtain an estimate of POM. The POM (mg) estimate is multiplied by a factor of 0.47 ± 0.01 (Dean 1974) to obtain an estimate of POC in the volume filtered (mg/L).

Lake water was filtered through Whatman, GF/F filters for DOC analysis. Filtered samples were stored at 4°C in HDPE bottles until analysis. A high temperature oxidation method was used to determine TDOC. After warming to room temperature, samples were analysed using a Shimadzu, Total Organic Carbon Analyzer (TOC-V_{CPH}). A calibration curve was prepared using potassium hydrogen phthalate (KHP) and machine performance was checked daily with a KHP NIST standard.

Lake water for coloured DOC (CDOC) analysis was filtered through a 0.22 μm Durapore (PVDF) membrane filter. Samples were stored in HDPE bottles at 4°C until analysis. Absorbance at 330 nm was used as an indication of the coloured DOC in sample

water {Kirk, 1994 #285}. Samples were analysed at 330 nm on a Spectronic, Genesys 5 spectrophotometer in a 5 cm path length, quartz cuvette.

3.3.4 Dissolved Carbohydrates

Filtered lake water (Whatman GF/F) was used to directly determine total dissolved monosaccharide (MONO), and after a hydrolysis step, total dissolved carbohydrates (TDCHO) via the TPTZ method (Myklestad et al. 1997). Total dissolved polysaccharide (POLY) was calculated as the difference between TDCHO and MONO. 1 ml of 0.7 mM potassium ferricyanide is added to a 1 ml sample (or hydrolysate) and placed in a boiling water bath for 10 minutes. While still hot, 1 ml of 2 mM ferric chloride and 2 ml of 2.5 mM TPTZ in 3 M acetic acid is added and mixed well on a vortex mixer. After 30 minutes, the absorbance was read at 595 nm on a Spectronic, Genesys 5 spectrophotometer using a 1 cm path length, disposable cuvette. All reagents were made fresh just before the analysis and all steps were performed in darkness as the analysis is light sensitive. Standards (66.66-266.66 μM) were made and analysed daily from individually frozen vials of the same 1000 ppm D-glucose stock solution. To hydrolyze the samples, HCl was added to a final concentration of 0.09 N before samples were sealed in glass ampoules and incubated at 150°C for 1 hour.

3.3.5 TEP Analysis

Semi-permanent slides were prepared in duplicate based on the method of Alldredge et al (1993) with few modifications. For details on slide preparation, TEP enumeration and calculation of TEP size distributions, please refer to the methods section 2.3.3.1 in Chapter 2. For details on the calculation of TEP-carbon, please refer to the methods section 2.4.1 in Chapter 2.

3.3.6 Bacteria

TEP-Attached and Free-Living Bacteria

To assess the number of TEP-attached and free-living bacteria, samples were prepared using the double staining method from Alldredge et al (1993). Samples were initially stained with DAPI (0.25 μg/ml final concentration) for at least 10 minutes before

being filtered through a 0.2 μm polycarbonate membrane filter, at a constant pressure below 150 mm Hg. Alcian blue was then added (0.5 ml) and drawn through immediately. Filtered material was transferred to a glass slide via the filter-transfer-freeze method (Hewes and Holm-Hansen 1983). A loop of gel (0.035 g/ml gelatine, 25% glycerine in distilled water) was placed over the filtered material while still frozen and left to solidify. Prepared slides were stored at -20°C in sealed bags until enumeration.

Enumeration of TEP-attached and free-living bacteria was performed at 1000x magnification (oil objective Leitz PL Apo) on a Leitz, Aristoplan, epifluorescence microscope configured with a 100-W HBO mercury bulb, and G340-380 excitation, RKP 400 dichromatic mirror, and LP430 suppression filter. By switching from bright field to epifluorescence, free-living bacteria could be defined on the basis that they were not associated with TEP. Conversely, TEP-attached bacteria are distinguished as being those bacteria which appear attached to TEP particles. An attempt was made to focus through the particle and count embedded bacteria, however, there was variability in the transparent nature of the TEP particles present. Therefore, it is possible that reported numbers of TEP-attached bacteria are an underestimate of actual abundances. For TEP particles that were completely transparent, it is possible that some free-living bacteria caught between the filter and particle were counted as attached bacteria. This error was always below 10% (maximum) and was not accounted for in calculated abundances. Free-living bacteria were counted in 10 fields. TEP-attached bacteria were counted on 20 different particles covering the size ranges present. The relationship between the number of bacteria attached per area TEP, and TEP particle size was applied to the size distribution of TEP particles, to get a total TEP-attached bacteria estimate.

Total Bacteria

Total bacteria samples were preserved in 1% glutaraldehyde, and stored in the dark at 4°C until slide preparation. Replicate slides were prepared based on the methods of Porter and Feig (1980) and Bae Yoon and Rosson (1990). A surfactant (Tween 80) was added to a final concentration of ~1%, vortexed and allowed to penetrate aggregates for at least 2 hours before being sonicated below 10 W for 30 seconds (Yoon and Rosson 1990). DAPI was then added to a final concentration of 10 μg/ml and let stand for at least

10 minutes. 1-2 ml sample aliquots were filtered on to phosphobuffered saline (PBS) conditioned, 0.2 μm, nucleopore filters, at a filter pressure of 178 mm Hg and was followed by a rinsing with PBS. Filtered bacteria were transferred to a glass slide using the filter-transfer-freeze method (Hewes and Holm-Hansen 1983) and covered with a loop of hot gel. Enumeration of bacteria was performed at 1000x magnification on a Leitz, Arisitocrat epifluorescence microscope. Bacteria were counted in 10 fields on each slide.

Other-Attached (OA) Bacteria

Other-attached bacteria abundances were inferred from the following equation:

TEP-Attached Bacteria = Total Bacteria – (Free-Living Bacteria + Other-Attached Bacteria).

3.3.7 Transfer Efficiency

Free-living and attached bacteria estimates are subject to error due to incomplete transfer of material using the filter-transfer-freeze method (Hewes and Holm-Hansen 1983). We found that our transfer efficiency for bacteria was >90%. We presume that the smaller the particle the more difficult the transfer and since the sizeable fraction of TEP is larger than most bacteria, we believe that transfer efficiencies for TEP would be even more complete. On this basis we make no corrections to bacteria or TEP abundances and view our estimates as methodologically robust.

3.3.8 Data Analysis

To test for differences in TEP abundance among lakes as well as the seasonal time effect within lakes, a randomized block design was performed with Lake set as a random (block) effect and Date as a fixed effect. This analysis gave the same result as a repeated measures ANOVA run in SAS. Correlation analysis was done by linear regression of x and y variables. Strength of the relationship was determined using the coefficient of determination (r^2) and significance was tested using ANOVA. Variables included in multiple linear regression analyses were chosen based on biological relevance and the strength of the relationship between the variable and TEP. Model performance was based on the r^2 and the inclusion of additional variables was based on significant increases to

the model's performance (r^2). All statistical analyses were performed in JMP 7 unless stated otherwise.

3.4 Results

3.4.1 Seasonal Variation in TEP Concentration

In 2006, seasonal TEP concentrations varied over two orders of magnitude from 296 ppm immediately after the flood (early June) to 5 ppm in late August (Table 3.1). After the initial sampling date TEP concentrations dropped rapidly in all lakes measured (Figure 3.2A). A randomized block design ANOVA was run to test for differences in TEP abundance over time. Early season river water input appears to influence TEP abundances heavily. We ran the ANOVA including and excluding the first sampling date to test whether there was an overall time effect (first sampling date included), and a time effect due to mainly in-lake processes (first sampling date excluded). There was a significant time effect for both iterations albeit at different significance levels ($F = 9.37$, $p < 0.0001$, $df = 10$ and $F = 2.88$, $p = 0.027$, $df = 9$ respectively).

TEP concentrations in 2007 ranged from 30 ppm to 7 ppm over a similar time frame as was surveyed in 2006 (Figure 3.2B and Table 3.2). The result of the randomized block design ANOVA testing for a time effect with the first sampling date included was significant ($F = 15.97$, $p = 0.0029$, $df = 3$) and insignificant with the first sampling date removed ($F = 2.47$, $p = 0.1998$, $df = 2$).

3.4.2 Inter-Lake Variation in TEP Concentration

In 2006, the mean TEP concentration in all lakes combined was 33.3 ppm with the first sampling date included and was 18.9 ppm without. Lakes 129, 56, and 520 had mean TEP concentrations of 48.8, 22.5, 28.5 ppm respectively with the first sampling date included and 24.1, 15.1, 17.5 ppm with the first sampling date excluded. For ease of comparison, descriptive statistics have been included in Table 3.1. The randomized block design ANOVA found no significant differences between lakes, with or without the first sampling date included, in either 2006 or 2007 seasons. Table 3.2 contains descriptive statistics for the 2007 data.

3.4.3 Size Distribution

We describe the size distribution of TEP using the spectral slope, b, from the size distribution equation (see methods). The spectral slope, b, ranged from 2.01 to 2.97 in lake 129, 1.99 to 3.44 in lake 56, and 2.02 to 3.42 in lake 520 (Figure 3.3) with a mean of 2.57 (SD = 0.31) , 2.84 (SD = 0.41), and 2.64 (SD = 0.36), respectively. The lower limit of the b-constant was essentially equal in the three lakes, while the maximum value for Lake 129 was lower than that of Lake 56 and 520. There was no evidence of a consistent seasonal trend in TEP size distributions in the lakes examined.

3.4.4 TEP Contributions to POC

TEP_{carbon} varied over an order of magnitude from 171 – 3242 μg C/L with an overall mean of 799 μg C/L (Figure 3.4A). As expected, the general seasonal trend follows that of TEP volume concentration with high TEP_{carbon} concentrations immediately after the flood which steadily decline towards the end of August. Mean TEP_{carbon} for Lakes 129, 56, and 520 were 1025, 790, and 561 μg C/L respectively. The mean contribution of TEP_{carbon} as a percentage of POC in the lakes was 83.7% and the seasonal variations are shown in Figure 3.4B. Individually, TEP contributed a mean 121.3, 69.0, and 69.9% of the POC found in Lakes 129, 56, and 520 respectively. Therefore, TEP contributions to POC appear to increase with increasing flood frequency.

3.4.5 Regression

Relevant chemical and biological variables were regressed on TEP to explore any relationships found within and between lakes. It is clear that the input of river water greatly impacted TEP concentrations in all lakes during the first week of sampling. This single data point is a result of a separate process (river water input) which was not quantified or had fallen to zero (high elevation lakes) and therefore the data point was not included in regression analyses. We take the remaining data points as representing in-lake processes which are influenced by river water input but not dictated by it. Results from the regression analysis are reported in Table 3.3.

Lake 129 (no-closure, high flood frequency)

The highly significant, positive relationship between TEP and TSS ($r^2 = 0.86$, $p<0.001$, df = 8) in Lake 129 is an indication of the impact of riverine inputs on TEP concentrations (Figure 3.5). CDOC, POC, and Chl a had equally strong relationships with TEP yielding coefficients of determination of 0.58, 0.57, and 0.56 respectively ($p<0.05$, df = 9, 8, and 9) (Figure 3.5). Each of these measures follow a similar trend to that of TSS.

Lake 56 (high-closure, low flood frequency)

POC ($r^2 = 0.62$, $p < 0.01$, df = 9), TDOC ($r^2 = 0.55$, $p < 0.05$, df = 9), and CDOC ($r^2 = 0.55$, $p < 0.05$, df = 9) had strong positive relationships with TEP concentrations in Lake 56 (Figure 3.6). POC is influenced by both early season inputs from the river, as well as algal abundances, as indicated by significant relationships between POC and TSS; and POC and Chl a concentrations ($r^2 = 0.41$ and $r^2 = 0.66$ respectively). The insignificant relationship between TEP concentration and TSS is an indication of diminished importance of river water for TEP abundances (Table 3.3).

Lake 520 (high-closure, low flood frequency, thermokarst activity)

TEP had a significant, positive relationship with total bacteria ($r^2 = 0.61$, $p < 0.05$, df = 8), OA-bacteria ($r^2 = 0.57$, $p < 0.05$, df = 8), and TEP-attached bacteria ($r^2 = 0.55$, $p < 0.05$, df = 8). This strong bacterial signal was not present in the other lakes. There was also a significant relationship between TEP concentration and CDOC (Figure 3.7) although it was the weakest of the three lakes ($r^2 = 0.40$).

3.4.6 Multiple Regression

After observing the strong river water influence on TEP concentration in low elevation Lake 129, the DOC influence in Lake 56, and the bacterial signal from Lake 520, we tested, post hoc, various models to see which variables are generally important for TEP concentration in Mackenzie Delta Lakes. Our original hypotheses and simple regression results guided our choice of predictor variables to include in the models. We chose to include POC due to its relation with both phytoplankton and river water. CDOC was included in the models because it is a general river water signal and was significantly

correlated with TEP in all lakes. TDOC was included because TEP-precursors are included in this measure, and because of the strong relationship found in Lake 56. Total bacteria was included because of the strong bacterial signal in Lake 520.

CDOC, on its own, predicted 48% of TEP variation in all lakes. Adding total bacteria to the model increased its r^2 significantly ($p = 0.0295$) from 0.48 to 0.62 ($p = 0.0001$). Adding OA-bacteria to CDOC instead of total bacteria resulted in marginal improvement ($r^2 = 0.63$, $p < 0.001$) as total and OA-bacteria are highly correlated ($r^2 = 0.94$, $p < 0.001$). Adding other variables to this model or CDOC did not explain any additional variation in TEP concentration.

3.4.7 Principal Components Analysis (PCA)

Since many of the variables that have a significant relationship with TEP concentration are correlated with one another (Table 3.4), we performed PCA to explore the possibility of reducing the large number of data variables to a small number of component variables that might explain TEP concentrations with greater parsimony. It appears that many component variables are needed to explain the bulk of the variance found in the data (Table 3.5) with the first component explaining just 32.84%. The first component had a significant relationship with TEP concentration ($r^2 = 0.57$, $p < 0.0001$, Figure 3.8), while subsequent components had insignificant relationships with TEP. Variables strongly correlated with the first principle component include TSS, POC, CDOC, polysaccharide, TEP-attached bacteria, OA-bacteria, and total bacteria. Our analysis included data from all the lakes combined as our sample size was too small to separate into individual lakes.

3.5 Discussion

3.5.1 Seasonal Variation in TEP Concentration

In 2006, a distinct seasonal pattern in TEP abundance (volume concentration) is apparent in all lakes from the initially high post-flood abundance and the subsequent gradual decrease towards late August. TEP abundances significantly decreased over the summer under scenarios where the highest TEP values (1st sampling date) were included in the statistical analysis and when they were excluded. We take this as evidence that both river influence and in-lake processes are important drivers of TEP abundances in Mackenzie Delta Lakes.

In 2007, the seasonal pattern of TEP abundances repeated itself with high post flood abundances quickly dropping before levelling off towards the end of August. Although the seasonal pattern of TEP abundances between 2006 and 2007 were similar, the actual 2007 TEP abundances were a magnitude lower immediately after the flood compared with samples from 2006. It is interesting that the late summer TEP abundances are so similar between years yet early season abundances are up to a magnitude different. Since samples from 2006 were prepared fresh and samples from 2007 were preserved in formalin, it is difficult to rule out the possibility that the 2007 samples were poorly preserved. However, according to Passow and Alldredge (1995b), TEP is preserved satisfactorily in formalin and the authors note that non-TEP artefacts may be present after preservation, indicating a tendency to overestimate TEP abundance. Slides prepared using formalin preserved TEP appeared no different than freshly prepared TEP slides. If TEP degradation had occurred, one would expect to find TEP particles in various stages of degradation. This was not evident from visual observation.

To attempt an explanation of the apparent inter-year variation in initial TEP concentration we must look at the potential causes. There are a number of possibilities. High under-ice TEP concentrations, high riverine TEP concentrations, an interaction between river water and lake water, or a flood stage effect are potential sources for such high TEP concentrations associated with the period immediately after the flood. Under ice TEP is an unlikely source for the observed TEP concentrations as values for Lake 520

were 138 ppm after being flooded in 2006 and were only 17 ppm in 2007, a low flood year in which the lake may not have been flooded. The sill elevation of Lake 520 is 4.93 m above sea level (asl). While the high water mark for 2007 was 5.02 m asl, the total flooding time for Lake 520 would have been <24 hours (Water Survey Canada). It is likely that Lake 520 received very little or no river water due to small differences in water levels between the gauging station located on East Channel and the inflow for Lake 520. Ice and snow may also play a role in blocking flood waters from entering the lake when potential flood times are so short and the differences between maximum flood height and sill elevation are so small (9 cm). We propose that the absence or small input of river water in Lake 520 during the spring of 2007 was the cause of lower TEP concentrations compared to Lakes 129 and 56 (Figure 3.2) which would have flooded. Increases in TEP concentration may occur in lakes containing high macrophyte abundances due to increases in DOC from decomposition over the ice covered season (Alber and Valiela 1994, Thornton 2004), however, these increases appear to be small. High riverine TEP concentrations may have caused the observed trends; however, this alone does not explain the apparent yearly variation in Lake 129. An interaction between lake water and river water occurring during the flood is possible, but it seems unlikely as the water in many of the lower elevation lakes would be essentially flushed and replaced by river water (Lesack et al. 1998), especially in high flood years as was observed in 2006. If the yearly variation observed in TEP concentrations is correct, then a combination of high riverine TEP concentration combined with a flood stage effect could account for our results. The 2006 spring flood was quite high (6.56 m asl) while the 2007 flood was comparatively low (5.02 m asl). We suggest that flood magnitude has an effect on riverine TEP concentration resulting in the great range of TEP values observed between the high and low flood years of 2006 and 2007 respectively. The reasons for such a great difference in TEP delivered via river water are unknown. Measuring TEP concentration at the outflow of Great Slave Lake would be a first step in further investigation of the phenomenon. An assessment of the potential for terrestrially derived DOC to form TEP would also be worth investigating as flood waters are mainly composed of runoff.

3.5.2 Inter-Lake Variation in TEP Concentration

We hypothesized that TEP concentration in Mackenzie Delta Lakes would increase with decreasing flood frequency (increasing sill elevation) due to gradients in chemical and biological variables. Our results show that there were no statistical differences in TEP concentration between the lakes in this study. This may be a result of the similar trophic status of the lakes examined as TEP concentration has been shown to increase with increasing trophic status (Carrias et al. 2002). Although not significantly different, the highest mean TEP concentration was found in a frequently flooded lake. This is the opposite result from what we predicted.

3.5.3 Size Distribution

We examined differences in size distribution within and between lakes using the constant, b (spectral slope), from the power law equation. When the spectral slope is equal to 3, each size interval contains an equal volume of TEP. As b decreases, the proportion of small particles decreases and as b increases the proportion of large particles decreases. Therefore, the spectral slope can be used as an indication of changes in TEP formation and removal processes.

Both Lake 56 and 520 experienced conditions where $b > 3$, indicating small particle enrichment such that small particles accounted for a greater volume than larger particles. This seems to be a rare occurrence; generally, it is believed that small particles contribute negligibly to TEP volume concentration estimates in comparison to larger particles (Passow 2002a). For Lake 520, the maximum b value occurred on the first sampling date (Figure 3.3), indicating an abundance of TEP-precursors forming small particles at rate exceeding removal via aggregation and sedimentation. A possible source of abundant TEP-precursors could be the recent input of fresh DOC from flood waters, although we did not observe such high values for the spectral slope in Lakes 56 or 129 during this time. Differences in aggregation rates between the lakes could account for the differences in spectral slope. This could be related to wind driven turbulence which would increase the number of particle collisions, resulting in aggregation where calmer waters could allow small TEP particle build-up. The surface area of Lake 56 is an order

of magnitude larger than Lake 520, and Lake 129 is 2 orders of magnitude larger, making Lake 56 and 129 more susceptible to wind mixing. This pattern fits with our results in that lake surface area in Lake 129 > Lake 56 > Lake 520 and the spectral slope in Lake 129 < Lake 56 < Lake 520 (see appendix A). However, specific examination of the relationship between wind mixing, lake surface area, in situ turbulence, and TEP size distributions would be required to confirm this hypothesis. In Lake 56, the maximum b value occurred on July 17 (Figure 3.3), but we have no evidence for a source of the TEP precursors required for an event such as this.

For each instance when b > 3, a sharp decline in the proportion of small particles present (decrease in b) was observed after two weeks (Figure 3.3). There was a similar rising and falling pattern in spectral slope observed for Lake 129 although its b value was never greater than 3. A switch from a small particle enriched state to depleted state could be the result of differential removal mechanisms such as rapid bacterial degradation of smaller particles or rapid removal via aggregation to larger particles. Although bacterial densities are always higher on smaller particles, there is no evidence to support the rapid bacterial degradation of the smaller TEP size classes, as the bacterial densities on small TEP were not substantially greater than average during the weeks in question (Chapter 4, Table 4.3). We did not measure bacterial activity, and it is possible that TEP-attached bacteria were more active during these time periods. As the spectral slope declines, there is a concomitant increase in TEP volume concentration, suggesting that small TEP aggregated to form larger TEP. This requires a return to more modest small particle formation rates, and is consistent with our understanding of the fractal nature of TEP. Our findings suggest that periods of small particle build-up will be terminated via rapid aggregation to large particles, indicated by a return to a small particle depleted state.

3.5.4 TEP Contributions to POC

An estimate of the proportion of POC contributed by TEP is an interesting proxy of the potential for TEP mediated carbon transfer between the dissolved and the particulate phases. Unfortunately, there is uncertainty associated with estimating the carbon content of TEP using equations from lab based studies. These uncertainties are discussed in Chapter 1 and elsewhere (Mari 1999, Engel and Passow 2001) and will not

be repeated here. We discuss TEP_{carbon} data as an exploration of the potential importance of TEP mediated carbon transfer from the DOC to POC in freshwater lakes with the acknowledgement that actual TEP_{carbon} values are likely lower than our estimates.

The importance of TEP for carbon cycling in freshwater ecosystems might be best described through comparison to the total amount of POC in situ. On average, 83.7% of POC was in the form of TEP in Mackenzie Delta Lakes. Depending on the rate of TEP turnover in these lakes, it is likely that the transfer of carbon from the DOC to POC via TEP formation is a significant pathway for carbon in this ecosystem, and may rival phytoplankton production. The ultimate fate of TEP_{carbon} is unknown but has the potential to contribute significantly to the sediments via sinking, to DIC and CO_2 flux via bacterial mineralization, and may represent a food source for protists (Tranvik et al. 1993) and zooplankton (Passow and Alldredge 1999, Ling and Alldredge 2003).

TEP turnover is likely to change predictably over time, depending on the prevailing conditions in the lakes. Using a steady state particle coagulation model, Mari and Burd (1998) found TEP turnover to be most sensitive to the concentration of non-TEP particles via the increased number of collisions and subsequent aggregation and removal. Their model estimated that TEP turnover rates range from $0.1 - 0.9\ d^{-1}$. Water residence time could also be a factor driving the rate of TEP turnover in Mackenzie Delta Lakes. Once a lake becomes disconnected from the river, it is generally not flooded again until the subsequent year, and sometimes longer, if it is an infrequently flooded lake. It has been shown that TEP turnover rates decrease as water residence time increases (Mari et al. 2007). This phenomenon is presumably due to prolonged bacterial degradation of organic matter in the system, and a subsequent decrease in the reactivity of organic matter. Since surface active colloids are responsible for TEP formation, a decrease in colloid reactivity would result in a decrease in TEP formation and turnover. With these two factors in mind, TEP turnover is likely to be high immediately after the flood due to higher particle concentrations and fresh sources of DOC, either directly from the river or indirectly through phytoplankton exudates. TEP turnover likely decreases as the summer progresses and particle concentrations decrease and water residence times increase, resulting in more refractory and less reactive DOC. If this in fact occurs, the carbon content of TEP within each size fraction will actually decrease as water residence time

increases, resulting in an overestimation of TEP_{carbon} (using the microscopic method) as the season progresses (Mari et al. 2007). The aging of DOC in these lakes could be mitigated to some extent through macroalgal (Alber and Valiela 1994, Thornton 2004), periphytic or epiphytic DOC inputs. We do not know to what extent this is true for Mackenzie Delta Lakes although there are substantial populations of these algal communities present. Some lakes also receive significant DOC contributions from non-algal macrophytes which have been shown to form aggregates under experimental conditions (Alber and Valiela 1994).

Lake 56 had a lower mean TEP volume concentration yet a higher TEP_{carbon} concentration when compared to that of Lake 520. Initially, this seems to be an anomaly. The result is partially due to the exclusion of the first sampling date from Lake 520 which was an obvious outlier, but we feel that more importantly, this demonstrates the effect of the TEP size distribution and the implications of the fractal nature of TEP. For example, two lakes may have the exact same TEP volume concentration but would have different TEP_{carbon} concentrations if their size distributions are different. This difference arises because smaller TEP are enriched in carbon compared to larger particles (Mari 1999). Therefore, a lake where TEP size distributions are skewed towards larger particles (low spectral slope) will have a lower value of TEP_{carbon} than a lake with a greater proportion of smaller particles (higher spectral slope), where TEP volume concentrations are equal.

3.5.5 Regression

The relationship between relevant chemical and biological variables and TEP concentration in individual lakes were not necessarily universal and suggest that the variables important for TEP concentration may be different depending on the prevailing conditions.

Lake 129 (low elevation, high flood frequency)

The strongest relationships found in Lake 129 were between TEP abundance and TSS, CDOC, POC, and Chl a (Table 3.3 and Figure 3.5). These variables are indicative of a strong river water signal in low elevation lakes, either through direct inputs, or an indirect mechanism such is nutrient input. TSS is the best indicator for TEP abundance

(based on r^2) when looking specifically at low elevation, Mackenzie Delta Lakes, due to their close relationship with the river compared to higher elevation lakes. It is unclear whether river water contributes TEP directly to the lake, although, as discussed earlier, this is a likely source for the high TEP abundances observed during the early summer. The influence of river water on this lake extends to other strong predictors of TEP abundance as well. This can be seen by the significant collinear relationships between TSS and CDOC, POC, and Chl a ($r^2 = 0.76$, $r^2 = 0.76$, and $r^2 = 0.73$ respectively).

Lake 56 (high elevation, low flood frequency)

Lake 56 exhibited a diminished river water influence on TEP concentration as shown by the insignificant relationship between TEP and TSS (Table 3.3). However, it is apparent that the initial river water input still influences TEP abundances, although it may occur through an indirect mechanism. POC was more heavily influenced by algal abundances than river water input, as shown by the stronger relationship between POC and Chl a concentration over POC and TSS concentration ($r^2 = 0.66$ and $r^2 = 0.41$ respectively), although both were significant. TEP concentration was also strongly related to TDOC and CDOC concentrations (Figure 3.6), which are a result of initial river water inputs, plus phytoplankton, epiphyton and macrophyte DOC contributions. From this we cautiously say that TEP concentrations in this lake are influenced by a mix of initial river water inputs plus DOC dynamics.

Lake 520 (high elevation, low flood frequency)

Short river-lake connection times for Lake 520 decrease the influence of river water inputs on TEP concentration as seen in the insignificant relationship between TEP and TSS (Table 3.3). The positive relationship between bacteria and TEP abundances indicates an increased importance of bacteria for TEP concentrations in higher elevation lakes that exhibit short connection times (Figure 3.7).

Factors Affecting TEP in Mackenzie Delta Lakes

We hypothesised that TEP would decrease with increasing TSS because TSS would aggregate with TEP, adding ballast and causing sedimentation. Our results showed that the relationship was actually opposite, with TEP concentrations generally increasing

with TSS concentration. While we are quite certain that TSS does cause some TEP sedimentation, there are a number of mitigating factors that go along with increases in TSS concentration in Mackenzie Delta Lakes. High concentrations of TSS are generally associated with the spring flood. Lakes that flood receive a significant input of sediments, nutrients, DOC, and it appears, TEP. Therefore, the initial input of river water drives (to varying extents) TEP abundances through direct (initial TEP input) and indirect (nutrient and DOC inputs for algal/bacterial growth) mechanisms. Our regression results indicate that the dominant factor for TEP concentration likely changes depending on the characteristics of the lake, and TSS concentration only gives an indication of general river water influence. We suggest that similarly to bacteria's influence on both TEP formation and degradation, suspended solids will affect both TEP formation and removal. As discussed above, the mechanism by which this would occur is through increasing particle concentrations which will increase the number of collisions with TEP, enhancing aggregation while at the same time enhancing removal through ballasting and subsequent sedimentation and particle scavenging. The balance between enhanced aggregation and removal basically describes suspended solids mediated TEP turnover, and will depend on the availability of TEP-precursors as well as turbulence in the water column which will, in turn, affect aggregation and aggregate settling efficiency. This balance is complex and deviations from steady state will result in either the accumulation or depletion of TEP in the water column.

The relationship between TEP and POC is a multi-faceted one, composed of TEP contributions to total POC, along with the relationship between POC and river water inputs (and associated TEP), phytoplankton abundance and growth phase, as well as the abundance and size distribution of TEP. Trends between each of these POC sources are expected to change across lakes of different elevations and even among lakes of similar elevation depending on prevailing conditions. For example, as discussed above, POC in Lake 129 is heavily influenced by river water through both direct inputs and nutrients which stimulate phytoplankton abundances. For the high elevation, Lake 56, there is a diminished effect of direct river water influence on POC and the input of nutrients and stimulation of phytoplankton populations causes a stronger relationship between POC and Chl a than POC and TSS. The weak and insignificant relationship between TEP and

POC in Lake 520 is surprising since TEP apparently contributes an average of ~70 % of all POC in the lake. The relationship could be obscured an overestimation of TEP_{carbon} and by complex nutrient recycling and carbon turnover, driven by the grazing activities of large zooplankton which are present in this lake due to the absence of fish (Riedel 2002).

In the case of TEP-attached bacteria, the relationship is an obvious result of bacterial colonization of TEP. However, the effect of OA- and total bacteria on TEP concentrations is complex due to direct and indirect bacterial influence on TEP production and decomposition. Bacteria can contribute to the production of TEP-precursors by stimulating the release of algal exopolymers through algal colonization and subsequent hydrolysis of mucus coatings (Smith et al. 1995). Bacterial colonization of algae on its own may also stimulate the production and release of exopolymers or TEP-precursors by algal cells (Passow 2002b). Grossart and Simon (2007) found that bacteria must be present for the formation of TEP which they hypothesize is either through bacterial modification of algal exuded DOC or decomposition of algae. Although production of TEP by bacteria through the release of precursors has been shown to contribute significantly to TEP abundances in a lab experiments (Stoderegger and Herndl 1999, Passow 2002b), in situ bacterial contributions are likely dominated by indirect stimulation of TEP formation (Grossart and Simon 2007, Sugimoto et al. 2007). Similarly, our results suggest that bacterial contributions to TEP abundances are greater than bacterial decomposition of TEP, however, the mechanism and pathway for this contribution is unclear. The only additional note we will make here is that the lower coefficient of determination for the relationship between TEP and total bacteria compared to TEP and OA-bacteria is likely due to the absence of a relationship between TEP and free-living bacteria (Table 3.3). This suggests that free-living bacteria have no net effect of TEP concentration. Perhaps the free-living bacterial population is in steady state with regard to TEP production and decomposition.

There are a number of possibilities why there is a strong and universal relationship between TEP and CDOC. Generally, the source of most CDOC is thought to be terrestrial. The strong relationship between CDOC and TEP abundances could be the result of terrestrially derived TEP-precursors. Alcian blue binds sulphated and carboxylated polysaccharides (Horobin 1988). While production of sulphated

polysaccharides is restricted to algal species (Kloareg and Quatrano 1988), all plants utilize the acidic polysaccharide, pectin in their cells walls (Scheller et al. 2007). Carboxyl groups associated with pectin should stain with alcian blue. Pectin is also known to form hydrogels in the presence of Ca^{2+} ions (Thakur et al. 1997), however, this has not been tested under in situ, lacustrine conditions. Orellana and Verdugo (2003) found that UV radiation inhibited the formation of self assembled gel particles. CDOC provides a natural sunscreen to aquatic environments and strongly absorbs UVA and UVB. As the summer proceeds, CDOC levels decline due to photobleaching (Gareis 2007) resulting in an increase in UV penetration. As CDOC decreases, UV radiation could inhibit TEP formation, especially in clear, high elevation lakes. Even though the previous two theoretical responses of TEP to CDOC are possible, it is also possible that CDOC merely describes the waning effect of river water inputs (which is the major pathway for CDOC entering lakes) and there may be little direct or indirect importance of CDOC for TEP abundance in Mackenzie Delta Lakes. It is clear that the relationship between CDOC and TEP requires further research and the potential for terrestrially derived DOC (specifically pectin) to contribute TEP-precursors should be explored experimentally. The potential for pectin-like acidic polysaccharides to cause the apparent overestimation of TEP using the spectrophotometric method could also be examined as the density of carboxyl groups associated with terrestrially derived polysaccharides may differ from the xanthan gum reference that is typically used.

The only significant relationship between TEP concentration and TDOC was found in Lake 56. As hypothesised, the relationship would be driven by the presence of TEP-precursors in the DOC for subsequent TEP formation. However, there was no apparent relationship in the other two lakes. Lake 129 and 520 never reached TDOC concentrations as high as Lake 56 so there may be a threshold at which the relationship between TDOC and TEP becomes apparent. Since there are many potential DOC sources, it is likely that DOC which will not aggregate into TEP, dominates TDOC in most lakes, effectively masking any relationship that might be present.

The relationship between Chl a and TEP is believed to be through the release of TEP precursors by phytoplankton, however the relationship is only significant in Lake 129 when lakes are analysed individually. There is actually a negative relationship

between TEP and Chl a in Lake 520, although it is an insignificant one (Table 3.3). It is clear that phytoplankton populations affect TEP abundances, but the relationship is not as simple as supplying TEP precursors and can obviously be affected by other processes such as bacterial abundances, and likely grazing rates.

We hypothesised that TEP concentrations would increase with carbohydrate concentrations. However, we found no significant relationships between TDCHO, dissolved mono-, or polysaccharide concentration when the lakes were analysed separately. There was a significant relationship between TEP and dissolved monosaccharide in all lakes combined, however we do not have any biological explanation for this pattern. TEP had a negative (although not significant) relationship with dissolved polysaccharides. Engel et al (2004) found that dissolved polysaccharide concentration decreased as TEP increased in a lab experiment. While this response could explain our findings, it is unlikely that weekly sampling would provide sufficient resolution to capture the effect.

We hypothesised that because autotrophic biomass increases with increasing sill elevation (decreasing flooding frequency), and autotrophic communities produce TEP-precursors, TEP concentration would increase with increasing sill elevation. We did not quantify the TEP-precursor contribution from macroalgae, periphyton, and epiphyton communities in Mackenzie Delta Lakes. However, it is apparent that these communities do not contribute enough TEP precursors to support higher TEP concentrations in high-sill lakes compared to low-sill lakes.

A single important variable will not predict TEP abundances for this ecosystem. This is likely due to a combination of the complexity of TEP formation and degradation in conjunction with the complexity of the ecosystem in which TEP is being formed and degraded. The differences in the relationships between the measured variables and TEP, in lakes of varying elevation, emphasizes the need for direct TEP measurements and highlights the difficulty associated with making generalizations among surficially similar aquatic ecosystems. In the case of TEP, it is not surprising when considering the potential sources and pathways for carbon through an aquatic ecosystem.

3.5.6 Multiple Regression

The exploratory regression of various models confirmed the strong river water influence, as well as bacterial influence, on TEP abundance in Mackenzie Delta Lakes. The best model included CDOC (river water influence) and total or OA-bacteria. The inclusion of TDOC in the model did not greatly improve the r^2, and suggests that the relationship between TDOC and TEP may be specific to Lake 56, or only occurs at higher TDOC concentrations as are found in Lake 56.

3.5.7 Principal Components Analysis (PCA)

The first component of the PCA explained relatively little of the variance in the data and was the only component that had a significant relationship with TEP. This component also had a significant relationship with a number of variables that are indicative of river water influence. It is likely that this component represents the impact of river water on these variables in the Mackenzie Delta. However, since so many components are needed to explain the bulk of the variance in the data, it appears that there are a great number of other processes controlling the variables in the data set. If the first component represents river water influence, then river water influence is the most important factor for TEP concentrations. This leaves a number of other processes that explain small portions of the remaining variance in TEP concentration. The resolution of this analysis may be limited by grouping all lakes together. If the analysis was performed with a greater sample size on individual lakes, we expect that the components would likely represent different processes and would be related to TEP concentration in different ways than observed here.

3.6 Conclusion

Mackenzie Delta Lakes exhibit a distinct seasonal pattern in TEP abundances driven by the major hydrologic event of the year, spring flood. TEP concentrations are high immediately after the flood, after which, concentrations drop rapidly towards the end of August. Initially, high TEP concentrations are evidently a result of river water influence, the specific mechanism of which is not known. Contrary to our hypotheses, we found that there was no statistical difference in TEP concentrations between lakes of different flooding frequency. Also contrary to our hypothesis, the low-closure lake (high flooding frequency) had the highest mean TEP concentration of the lakes surveyed.

The seasonal trend of high post flood TEP concentrations (relative to late summer) was present in both 2006 and 2007. However, yearly variation was high (an order of magnitude) for post flood TEP concentrations and similar for the rest of the summer. We propose that flood magnitude mediates post flood TEP concentrations, while in-lake processes mediate summer TEP concentrations.

TEP size distributions indicated that rapid, small particle build-up can occur such that volume concentrations are influenced more by small particles than large particles, which seems to be a rare occurrence. Small particle build-up (spectral slope, $b > 3$) was terminated within two weeks through aggregation, but not necessarily removal.

The importance of different variables for TEP concentration changes depending on the biological and chemical conditions present in the lake. TEP concentrations in low-closure lakes with high flooding frequency are closely related to river water inputs. In high-closure lakes with low flooding frequency, river water inputs had a diminished effect on TEP concentration with DOC or bacteria being of greater importance. Multiple regression confirmed the importance of river water, DOC, and bacterial influence on TEP concentration as the model containing CDOC plus total bacteria explained the greatest amount of variation (62%) in TEP concentration for all Mackenzie Delta Lakes surveyed. Principle component analysis also suggested that river water influence was the main process controlling TEP concentrations.

Our results show that mean TEP contributions to POC increased with increasing flooding frequency. TEP_{carbon} contributes a large proportion (mean 83.7%) of POC in Mackenzie Delta Lakes. While this estimate is likely an overestimate, it is an indication that TEP represents a significant pathway for the transfer of carbon from dissolved to particulate phase. TEP turnover rates need to be determined to estimate the total amount of carbon cycling through TEP, while information about the fate of TEP should also be determined.

3.7 Figures

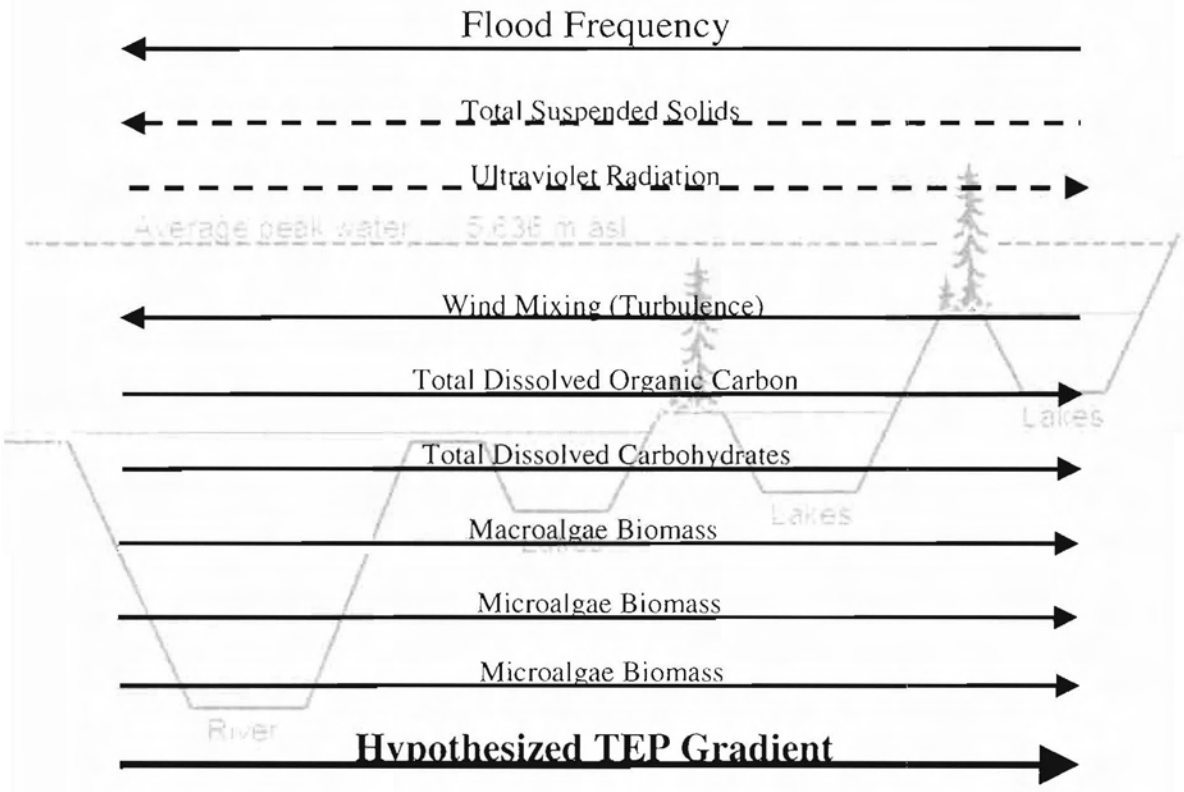


Figure 3.1 **General trends in physical, chemical, and biological variables are super-imposed on a graphical depiction of lakes of differing sill elevation. The arrow depicts the increasing direction of the trend. Solid lines represent a positive effect on TEP concentration while perforated lines represent a negative effect on TEP concentration. The result is a hypothesized TEP gradient that increases with decreasing flood frequency (sill elevation).**

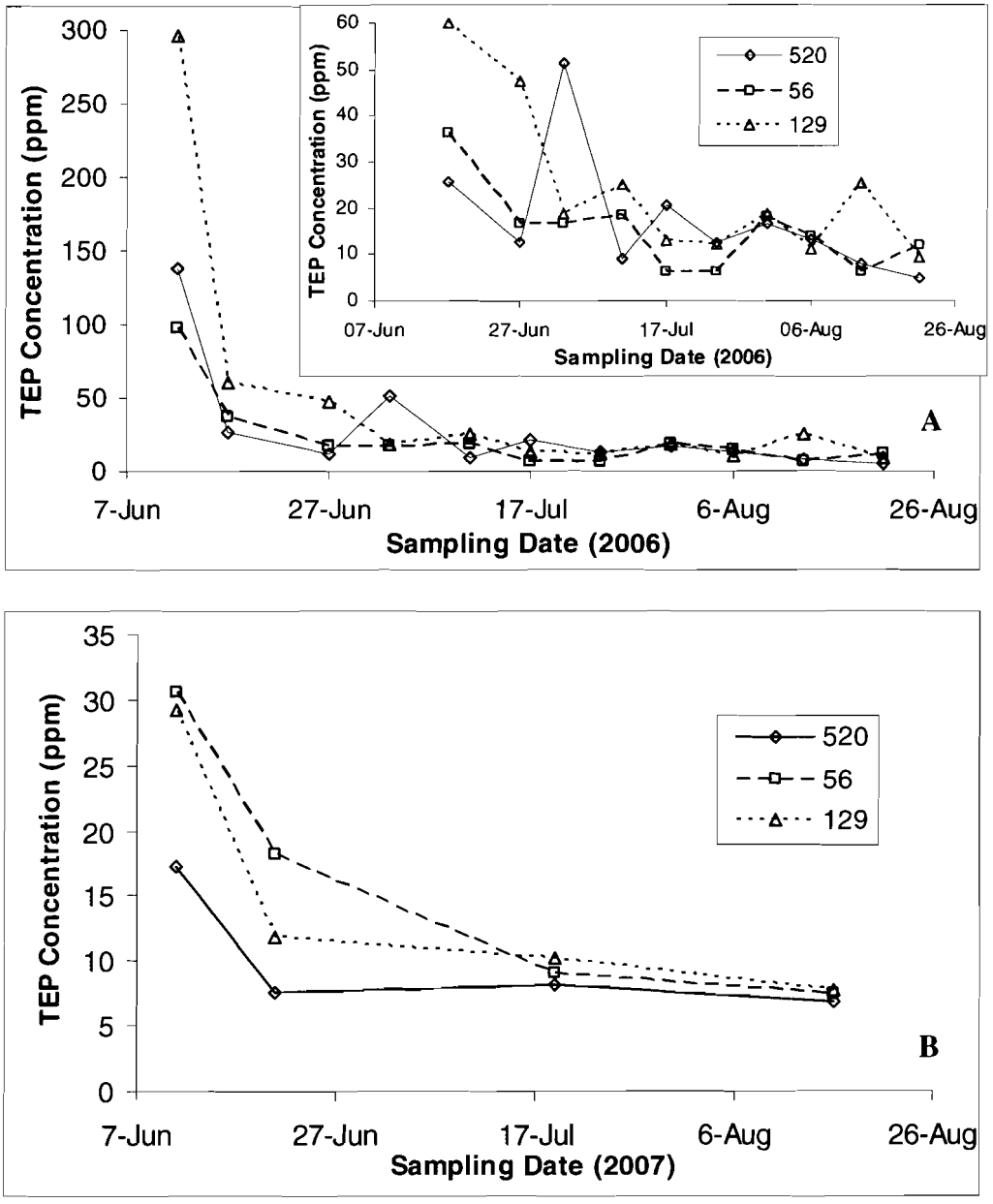
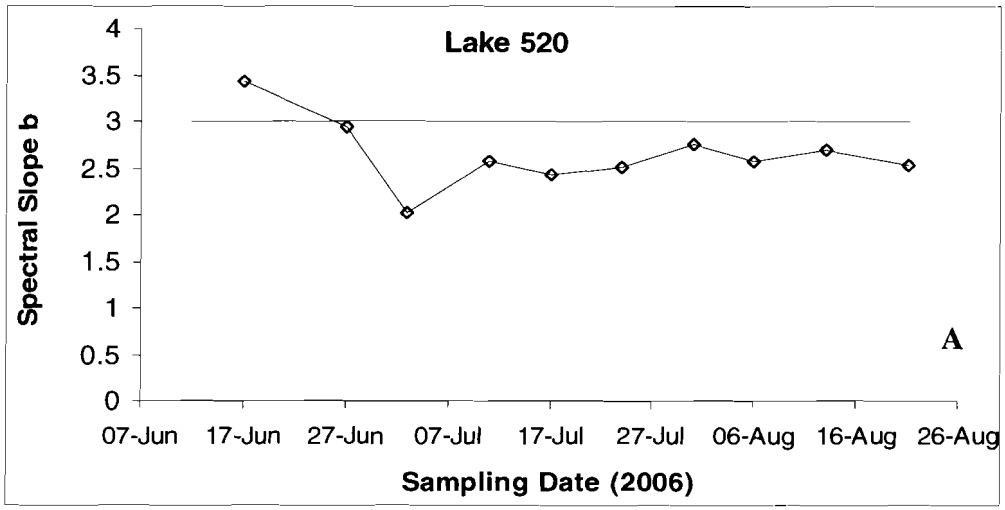
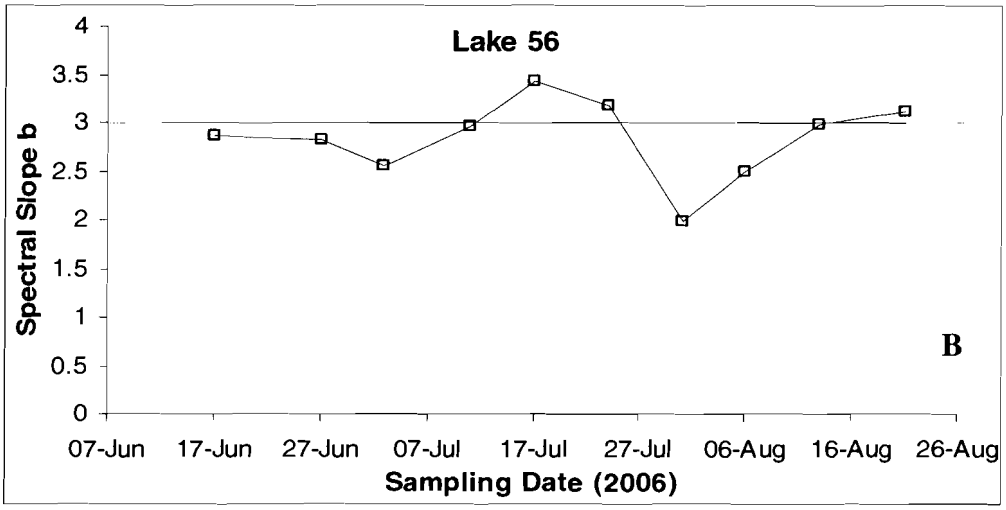


Figure 3.2 **Seasonal TEP abundances for the open water seasons of A) 2006 and B) 2007. Inset in A) shows greater detail in TEP concentration during the 2006 season.**





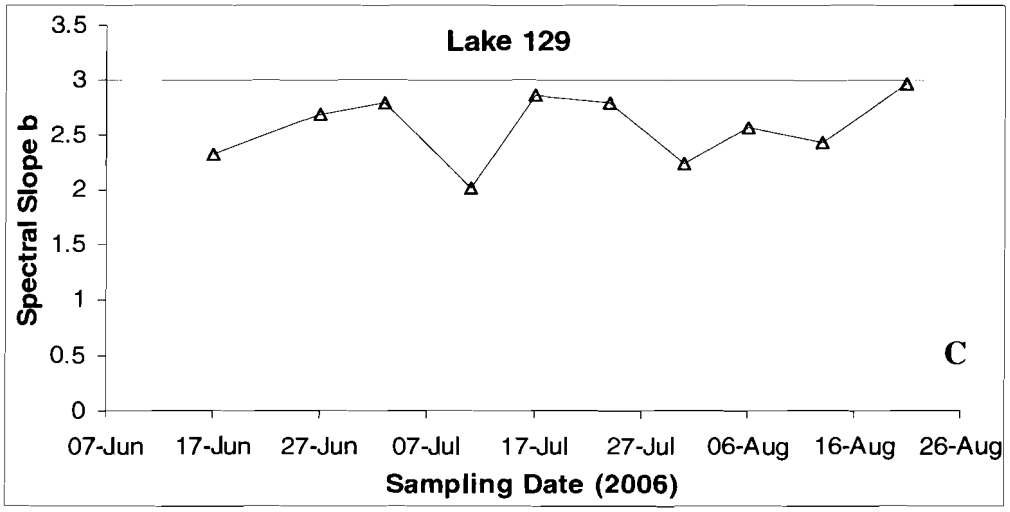


Figure 3.3 **Seasonal changes in the spectral slope (size distribution constant, b) for Lakes A) 520, B) 56, and C) 129. A line has been super-imposed at a spectral slope equal to 3, at which all particle sizes contribute equally to total volume concentration. Above this line, smaller particles dominate the volume concentration while below, large particles dominate.**

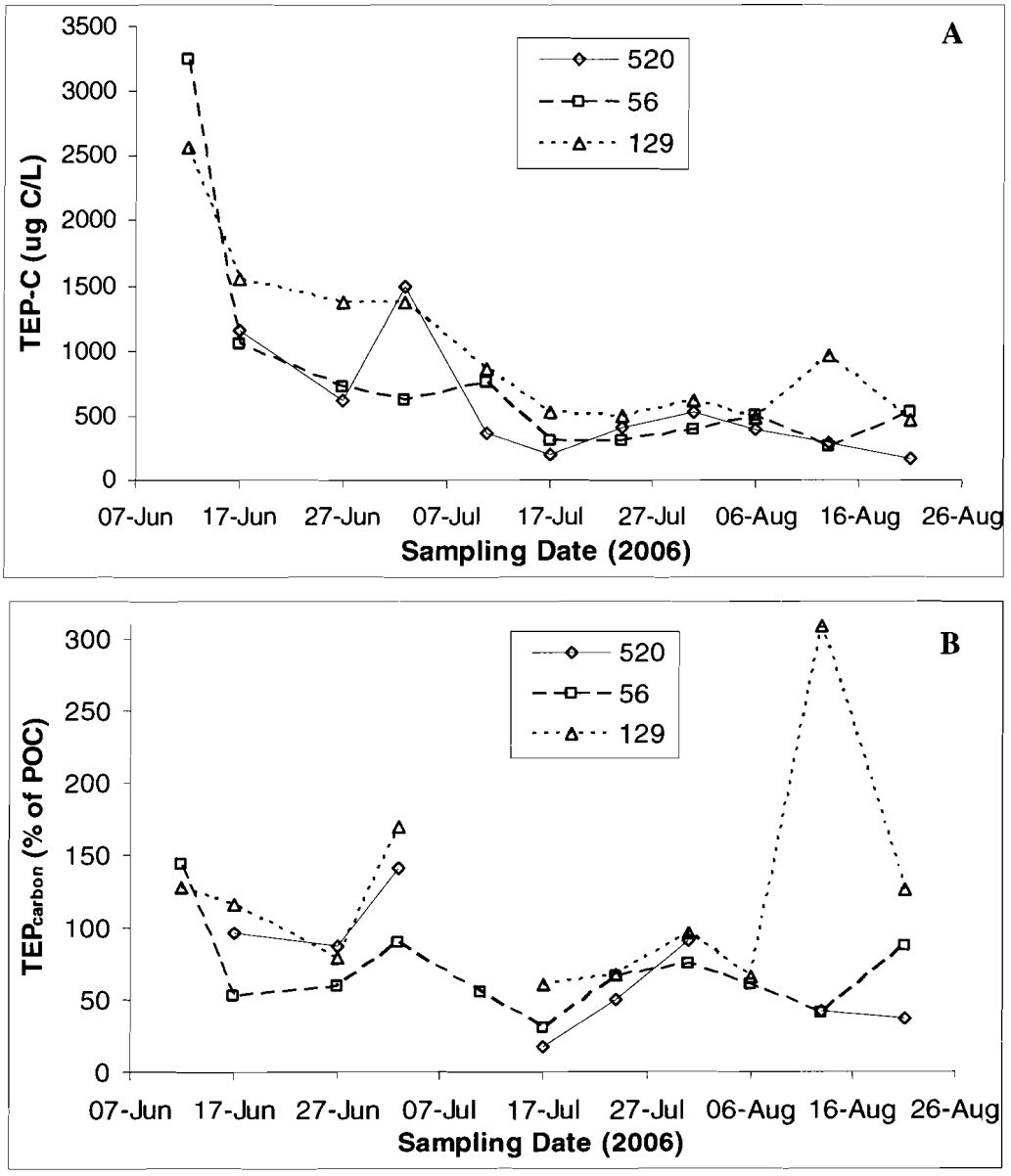


Figure 3.4 **Seasonal changes in (A) TEP_{carbon} (µg C/L) and (B) the % contribution of TEP to POC for Lakes 520, 56, and 129.**

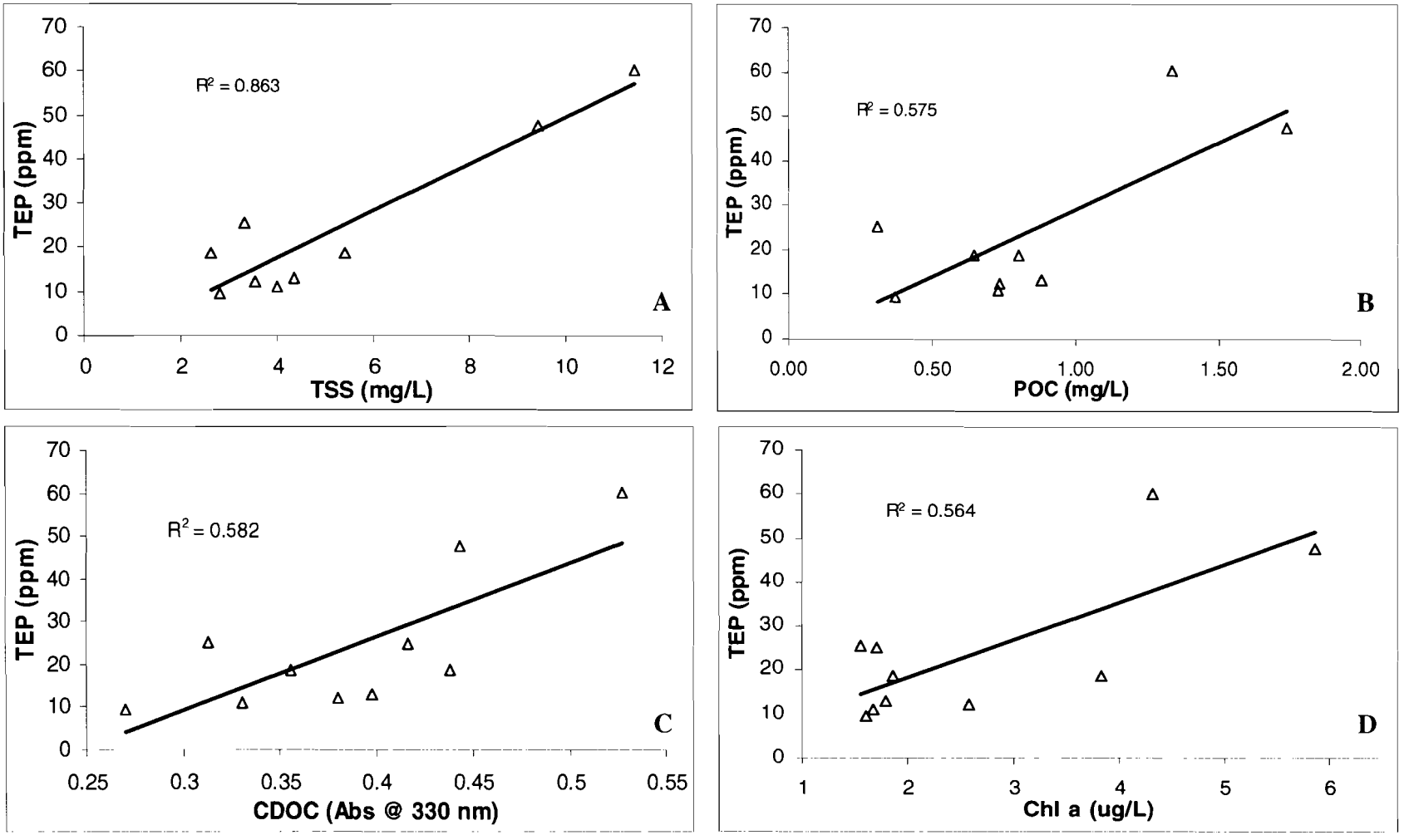


Figure 3.5 Plots show significant results of the linear regression of TEP abundance (ppm) with A) TSS (mg/L), B) POC (mg/L), C) CDOC (absorbance at 330 nm), and D) Chl a (μg/L) in Lake 129. The coefficient of determination is given for each relationship.

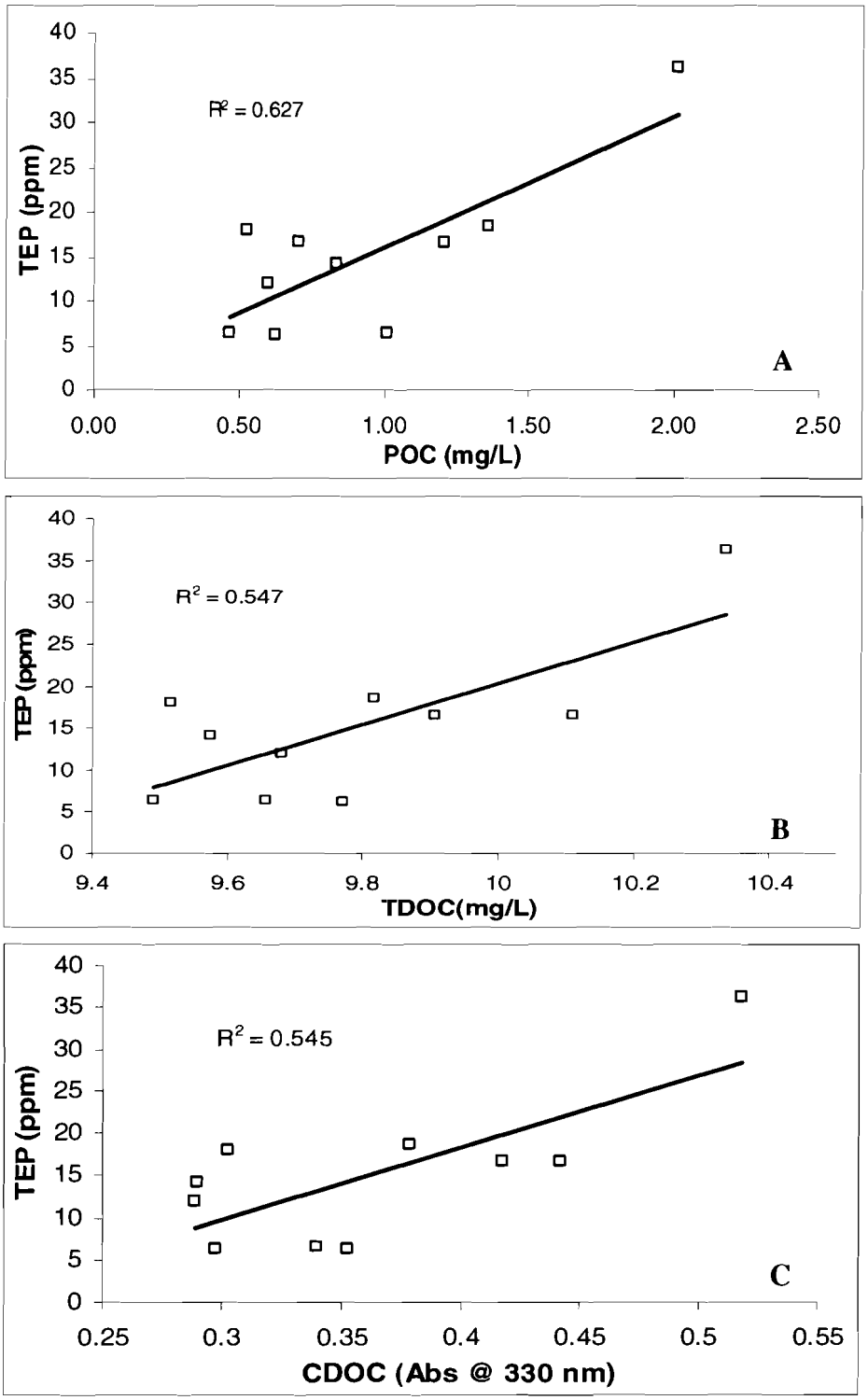


Figure 3.6 **Plots show significant results of the linear regression of TEP abundance (ppm) with A) POC (mg/L), B) TDOC (mg/L), and C) CDOC (absorbance at 330 nm) in Lake 56. The coefficient of determination is given for each relationship.**

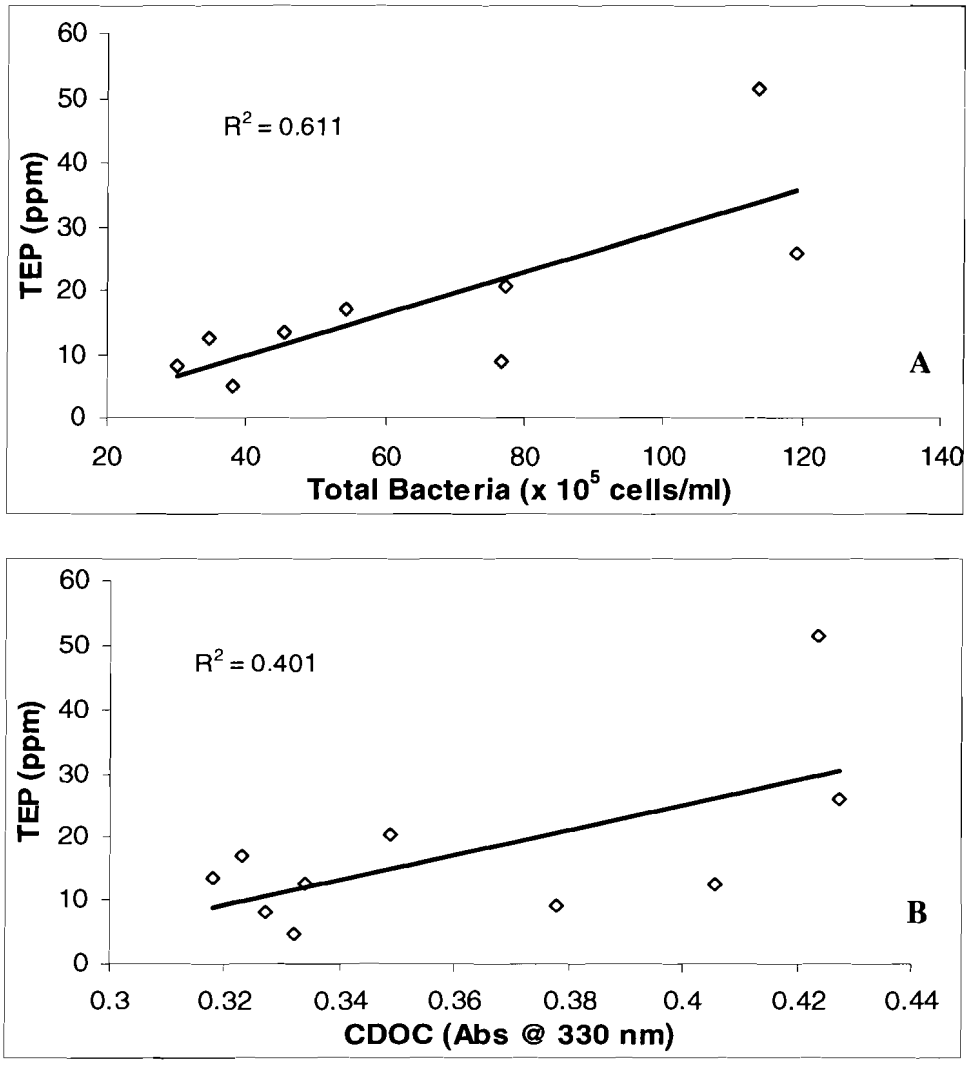


Figure 3.7 **Plots show significant results of the linear regression of TEP abundance (ppm) with A) Total Bacteria (# x10^5 cells/ml) and B) CDOC (absorbance at 330 nm) in Lake 520. The coefficient of determination is given for each relationship.**

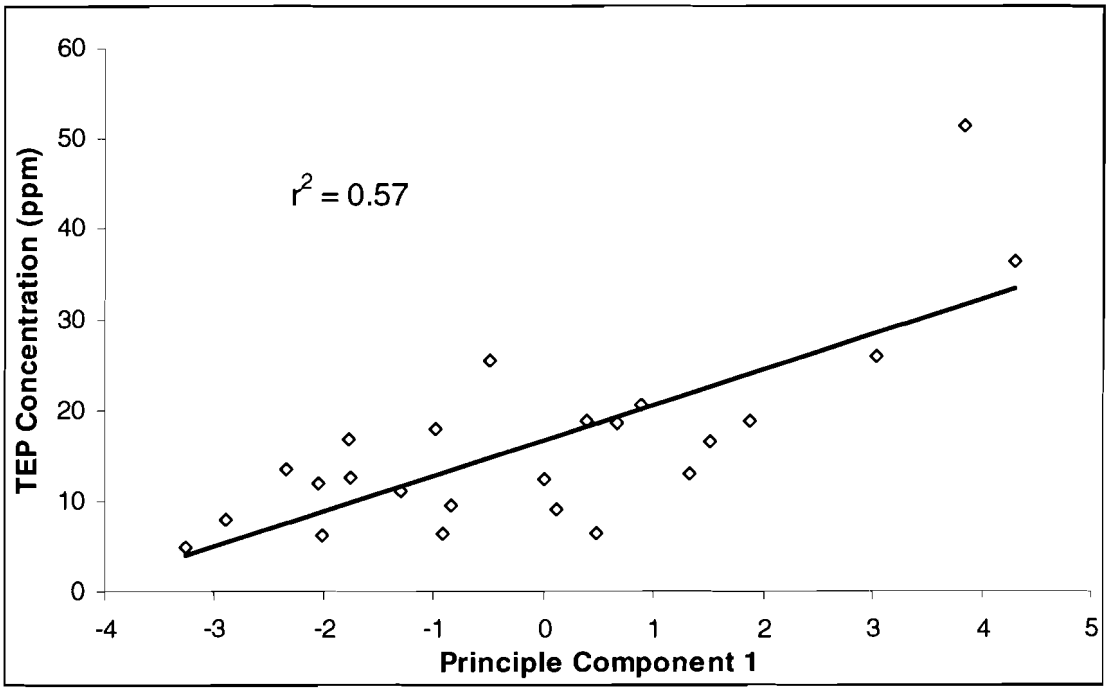


Figure 3.8 **Plot shows the significant relationship ($p < 0.0001$) between TEP concentration (ppm) and the first principle component from the PCA. The coefficient of determination is given.**

3.8 Tables

Table 3.1 Descriptive statistics for TEP abundances (ppm) during the summer of 2006.

	N	Mean	SD	Min	Max
First sample date included					
All Lakes	33	33.28	54.61	4.81	295.82
129	11	48.84	48.84	9.43	295.82
56	11	22.54	22.54	6.23	96.81
520	11	28.46	28.46	4.81	137.99
First sample date excluded					
All Lakes	30	18.92	13.55	4.81	60.12
129	10	24.14	16.85	9.43	60.12
56	10	15.11	8.90	6.23	36.22
520	10	17.50	13.40	4.81	51.36

Table 3.2 Descriptive statistics for TEP abundances (ppm) during the summer of 2007.

	N	Mean	SD	Min	Max
First sample date included					
All Lakes	12	13.65	8.48	6.76	30.61
129	4	14.75	9.82	7.73	29.26
56	4	16.26	10.67	7.38	30.61
520	4	9.95	4.95	6.76	17.32
First sample date excluded					
All Lakes	9	9.63	3.55	6.76	18.09
129	3	9.92	2.07	7.73	11.85
56	3	11.48	5.78	7.38	18.09
520	3	7.49	0.68	6.76	8.11

Table 3.3 **Coefficient of determination for variables plotted against TEP abundance. A + sign indicates a positive relationship, a – sign indicates a negative relationship. *, **, ***, indicates significance at the alpha = 0.05, 0.01, 0.001 levels respectively.**

Variable	520	56	129	All Lakes
TDOC	- 0.19	+ 0.55*	+ 0.02	- 0.02
TDCHO	- 0.01	0.00	+ 0.36	+ 0.02
Monosaccharide	+ 0.14	+ 0.04	+ 0.33	+ 0.18*
Polysaccharide	- 0.34	- 0.21	+ 0.06	- 0.11
CDOC	+ 0.40*	+ 0.55*	+ 0.58*	+ 0.48***
Chl a	- 0.25	+ 0.14	+ 0.56*	+ 0.21*
TSS	+ 0.31	+ 0.04	+ 0.86***	+ 0.40***
POC	+ 0.43	+ 0.62**	+ 0.57*	+ 0.40***
Total Bacteria	+ 0.61*	+ 0.03	0.00	+ 0.39***
Free-living Bacteria	+ 0.09	- 0.05	+ 0.17	0.00
TEP-Attached Bacteria	+ 0.55*	+ 0.01	+ 0.15	+ 0.20*
Other Attached-Bacteria	+ 0.57*	+ 0.17	- 0.06	+ 0.51***

Table 3.4 Correlation matrix for variables included in the principle components analysis.

Variable	TEP	TSS	POC	CDOC	Chl a	TDOC	TDCHO	Mono	Poly	Free Living Bacteria	Attached Bacteria	Residual Bacteria	Total Bacteria
TEP		0.33	0.53	0.61	0.09	-0.10	-0.07	0.27	-0.47	-0.05	0.49	0.53	0.53
TSS	0.33		0.69	0.48	0.13	0.15	-0.24	-0.05	-0.31	0.02	0.18	0.23	0.24
POC	0.53	0.69		0.78	0.37	0.32	-0.23	-0.04	-0.31	0.18	0.16	0.24	0.29
CDOC	0.61	0.48	0.78		0.50	0.19	-0.17	0.21	-0.55	0.23	0.31	0.46	0.52
Chl a	0.09	0.13	0.37	0.50		0.12	-0.01	0.10	-0.14	-0.25	-0.11	-0.18	-0.25
TDOC	-0.10	0.15	0.32	0.19	0.12		0.33	0.04	0.46	0.46	-0.07	-0.01	0.10
TDCHO	-0.07	-0.24	-0.23	-0.17	-0.01	0.33		0.78	0.55	0.27	0.08	-0.32	-0.20
Mono	0.27	-0.05	-0.04	0.21	0.10	0.04	0.78		-0.10	0.28	0.18	0.03	0.13
Poly	-0.47	-0.31	-0.31	-0.55	-0.14	0.46	0.55	-0.10		0.05	-0.12	-0.55	-0.49
Free Living Bacteria	-0.05	0.02	0.18	0.23	-0.25	0.46	0.27	0.28	0.05		0.08	0.09	0.37
Attached Bacteria	0.49	0.18	0.16	0.31	-0.11	-0.07	0.08	0.18	-0.12	0.08		0.28	0.43
Residual Bacteria	0.53	0.23	0.24	0.46	-0.18	-0.01	-0.32	0.03	-0.55	0.09	0.28		0.95
Total Bacteria	0.53	0.24	0.29	0.52	-0.25	0.10	-0.20	0.13	-0.49	0.37	0.43	0.95	

Table 3.5 **Results of the principle component analysis for the 13 variables shown in the correlation matrix (Table 3.4).**

Principal Component	Eigenvalue	% of Variance Explained	Cumulative % of Variance Explained
1	4.27	32.84	32.84
2	2.32	17.85	50.69
3	1.94	14.92	65.62
4	1.51	11.61	77.22
5	0.95	7.31	84.53
6	0.71	5.44	89.97
7	0.58	4.49	94.47
8	0.40	3.07	97.54
9	0.15	1.14	98.68
10	0.10	0.81	99.49
11	0.07	0.51	100.00
12	0.00	0.00	100.00
13	-0.00	-0.00	100.00

3.9 References

Alber, M. and I. Valiela. 1994. Production of microbial organic aggregates from macrophyte-derived dissolved organic material. Limnology and Oceanography **39**:37-50.

Alldredge, A. L., U. Passow, and B. E. Logan. 1993. The abundance and significance of a class of large, transparent organic particles in the ocean. Deep-Sea Research Part I-Oceanographic Research Papers **40**:1131-1140.

Carrias, J. F., J. P. Serre, T. Sime-Ngando, and C. Amblard. 2002. Distribution, size, and bacterial colonization of pico- and nano-detrital organic particles (DOP) in two lakes of different trophic status. Limnology and Oceanography **47**:1202-1209.

Carson, M. A., F. M. Conly, and J. N. Jasper. 1999. Riverine sediment balance of the Mackenzie Delta, Northwest Territories, Canada. Hydrological Processes **13**:2499-2518.

Chin, W. C., M. V. Orellana, and P. Verdugo. 1998. Spontaneous assembly of marine dissolved organic matter into polymer gels. Nature **391**:568-572.

Dean, W. E. 1974. Determination of carbonate and organic-matter in calcareous sediments and sedimentary-rocks by loss on ignition - Comparison with other methods. Journal of Sedimentary Petrology **44**:242-248.

Decho, A. W. 1990. Microbial exopolymer secretions in ocean environments - Their role(s) in food webs and marine processes. Oceanography and Marine Biology **28**:73-153.

Emmerton, C. A., L. F. W. Lesack, and P. Marsh. 2007. Lake abundance, potential water storage, and habitat distribution in the Mackenzie River Delta, western Canadian Arctic. Water Resources Research **43**.

Emmerton, C. A., L. F. W. Lesack, and W. F. Vincent. 2008. Mackenzie River nutrient delivery to the Arctic Ocean and effects of the Mackenzie Delta during open water conditions. Global Biogeochemical Cycles **22**.

Engel, A. and U. Passow. 2001. Carbon and nitrogen content of transparent exopolymer particles (TEP) in relation to their alcian blue adsorption. Marine Ecology-Progress Series **219**:1-10.

Engel, A., S. Thoms, U. Riebesell, E. Rochelle-Newall, and I. Zondervan. 2004. Polysaccharide aggregation as a potential sink of marine dissolved organic carbon. Nature **428**:929-932.

Gareis, J. 2007. Underwater irradiance attenuation and photobleaching of chromophoric dissolved organic matter in shallow arctic lakes of the Mackenzie Delta, NWT. Simaon Fraser University, Burnaby.

Grossart, H. P. and M. Simon. 2007. Interactions of planktonic algae and bacteria: effects on algal growth and organic matter dynamics. Aquatic Microbial Ecology **47**:163-176.

Hewes, C. D. and O. Holm-Hansen. 1983. A method for recovering nanoplankton from filters for identification with the microscope - The filter-transfer-freeze (ftf) technique. Limnology and Oceanography **28**:389-394.

Horobin, R. W. 1988. Understanding histochemistry. Selection, evaluation and design of biological stains. Wiley.

Kirk, J. 1994. Light and photosynthesis in aquatic ecosystems. Second edition. Cambridge University Press, Cambridge, UK.

Kloareg, B. and R. S. Quatrano. 1988. Structure of the cell-walls of marine-algae and ecophysiological functions of the matrix polysaccharides. Oceanography and Marine Biology **26**:259-315.

Leppard, G. G. 1995. The characterization of algal and microbial mucilages and their aggregates in aquatic ecosystems. Science of the Total Environment **165**:103-131.

Lesack, L. F. W., P. Marsh, and R. E. Hecky. 1998. Spatial and temporal dynamics of major solute chemistry among Mackenzie Delta lakes. Limnology and Oceanography **43**:1530-1543.

Ling, S. C. and A. L. Alldredge. 2003. Does the marine copepod Calanus pacificus consume transparent exopolymer particles (TEP)? Journal of Plankton Research **25**:507-515.

Long, R. A. and F. Azam. 1996. Abundant protein-containing particles in the sea. Aquatic Microbial Ecology **10**:213-221.

Mackay, J. R. 1963. The Mackenzie Delta Area, NWT, Canada. Canadian Department of Mines and Technical Surveys, Geographical Branch **Memoir No. 8**:202pp.

Mari, X. 1999. Carbon content and C:N ratio of transparent exopolymeric particles (TEP) produced by bubbling exudates of diatoms. Marine Ecology-Progress Series **183**:59-71.

Mari, X. and A. Burd. 1998. Seasonal size spectra of transparent exopolymeric particles (TEP) in a coastal sea and comparison with those predicted using coagulation theory. Marine Ecology-Progress Series **163**:63-76.

Mari, X., E. Rochelle-Newall, J. P. Torreton, O. Pringault, A. Jouon, and C. Migon. 2007. Water residence time: A regulatory factor of the DOM to POM transfer efficiency. Limnology and Oceanography **52**:808-819.

Marsh, P. and M. Hey. 1989. The flooding hydrology of Mackenzie Delta Lakes near Inuvik, NWT, Canada. Arctic **42**:41-49.

Marsh, P. and M. Hey. 1994. Analysis of spring highwater events in the Mackenzie Delta and implications for lake and terrestrial flooding. Geografiska Annaler Series a-Physical Geography **76**:221-234.

Marsh, P., L. F. W. Lesack, and A. Roberts. 1999. Lake sedimentation in the Mackenzie Delta, NWT. Hydrological Processes **13**:2519-+.

Mostajir, B., J. R. Dolan, and F. Rassoulzadegan. 1995. A simple method for the quantification of a class of labile marine pico- and nano-sized detritus: DAPI Yellow Particles (DYP). Aquatic Microbial Ecology **9**:259-266.

Myklestad, S. M. 1995. Release of extracellular products by phytoplankton with special emphasis on polysaccharides. Science of the Total Environment **165**:155-164.

Myklestad, S. M., E. Skanoy, and S. Hestmann. 1997. A sensitive and rapid method for analysis of dissolved mono- and polysaccharides in seawater. Marine Chemistry **56**:279-286.

Orellana, M. V. and P. Verdugo. 2003. Ultraviolet radiation blocks the organic carbon exchange between the dissolved phase and the gel phase in the ocean. Limnology and Oceanography **48**:1618-1623.

Passow, U. 2002a. Transparent exopolymer particles (TEP) in aquatic environments. Progress in Oceanography **55**:287-333.

Passow, U. 2002b. Production of transparent exopolymer particles (TEP) by phyto- and bacterioplankton. Marine Ecology-Progress Series **236**:1-12.

Passow, U. and A. L. Alldredge. 1995b. A dye-binding assay for the spectrophotometric measurement of transparent exopolymer particles (TEP). Limnology and Oceanography **40**:1326-1335.

Passow, U. and A. L. Alldredge. 1999. Do transparent exopolymer particles (TEP) inhibit grazing by the euphausiid Euphausia pacifica? Journal of Plankton Research **21**:2203-2217.

Porter, K. G. and Y. S. Feig. 1980. The use of DAPI for identifying and counting aquatic microflora. Limnology and Oceanography **25**:943-948.

Radic, T., I. Ivancic, D. Fuks, and J. Radic. 2006. Marine bacterioplankton production of polysaccharidic and proteinaceous particles under different nutrient regimes. Fems Microbiology Ecology **58**:333-342.

Radic, T., J. Radic, M. Najdek, D. Fuks, and M. Blazina. 2003. Potential of the northern Adriatic bacterioplankton for the production of transparent exopolyrner particles. Periodicum Biologorum **105**:381-387.

Riedel, A. 2002. Zooplankton composition and control of heterotrophic flagellates among lakes of the Mackenzie Delta. Simon Fraser University, Burnaby.

Scheller, H. V., J. K. Jensen, S. O. Sorensen, J. Harholt, and N. Geshi. 2007. Biosynthesis of pectin. Physiologia Plantarum **129**:283-295.

Smith, D. C., G. F. Steward, R. A. Long, and F. Azam. 1995. Bacterial mediation of carbon fluxes during a diatom bloom in a mesocosm. Deep-Sea Research Part Ii-Topical Studies in Oceanography **42**:75-97.

Squires, M. M. and L. F. W. Lesack. 2003a. Spatial and temporal patterns of light attenuation among lakes of the Mackenzie Delta. Freshwater Biology **48**:1-20.

Squires, M. M., L. F. W. Lesack, and D. Huebert. 2002a. The influence of water transparency on the distribution and abundance of macrophytes among lakes of the Mackenzie Delta, Western Canadian Arctic. Freshwater Biology **47**:2123-2135.

Stoderegger, K. E. and G. J. Herndl. 1999. Production of exopolymer particles by marine bacterioplankton under contrasting turbulence conditions. Marine Ecology-Progress Series **189**:9-16.

Sugimoto, K., H. Fukuda, M. A. Baki, and I. Koike. 2007. Bacterial contributions to formation of transparent exopolymer particles (TEP) and seasonal trends in coastal waters of Sagami Bay, Japan. Aquatic Microbial Ecology **46**:31-41.

Thakur, B. R., R. K. Singh, and A. K. Handa. 1997. Chemistry and uses of pectin - A review. Critical Reviews in Food Science and Nutrition **37**:47-73.

Thornton, D. C. O. 2004. Formation of transparent exopolymeric particles (TEP) from macroalgal detritus. Marine Ecology-Progress Series **282**:1-12.

Tranvik, L. J., E. B. Sherr, and B. F. Sherr. 1993. Uptake and utilization of colloidal DOM by heterotrophic flagellates in seawater. Marine Ecology-Progress Series **92**:301-309.

Wotton, R. S. 2005. The essential role of exopolymers (EPS) in aquatic systems. Pages 57-94 Oceanography and Marine Biology: An Annual Review, Vol 42.

Yoon, W. B. and R. A. Rosson. 1990. Improved method of enumeration of attached bacteria for study of fluctuation in the abundance of attached and free-living bacteria in response to diel variation in seawater turbidity. Applied and Environmental Microbiology **56**:595-600.

4 FREE-LIVING, TEP-ATTACHED, AND OTHER-ATTACHED BACTERIA AMONG MACKENZIE DELTA LAKES

4.1 Abstract

During June through August 2006, we conducted weekly surveys of the abundance of free-living and attached bacteria in the water columns of 3 Mackenzie Delta Lakes of differing flooding frequency. A bacteria budget was constructed to assess the relative importance of free-living, TEP-attached, and other-attached (OA) bacteria. We tracked seasonal and inter-lake changes in budget components as well as TEP-attached bacterial densities. Our results indicate that total bacterial abundances were high after the flood, and showed a significant decrease towards the end of August. Total bacterial abundances were high and ranged from ~10^6 to 10^7 cells/ml. On average free-living bacteria contributed 13.9% of total bacterial abundance, TEP-attached bacteria contributed 9.6%, and OA-bacteria contributed 76.5 %. TEP-attached bacterial densities were significantly higher in lakes of low flood frequency (high elevation) and densities increased over the summer, although not significantly in all TEP size intervals. Significant negative correlation between TEP-attached bacterial density and particulate matter, including TEP, suggest that TEP-attached bacterial density is related to the abundance of favourable colonization sites.

4.2 Introduction

In the aquatic sciences there are currently shifting views of how we perceive the environment in which microbes exist. The simplistic view of a homogenous environment where microbes, chemicals, and substrates are distributed evenly is outdated. However, in practice we continue to analyse the microbial community in conjunction with "bulk" water column characteristics as a result of methodological constraints or the time consuming nature of separation processes. Accumulating evidence shows that microbes encounter a complex, heterogenous environment of chemical gradients and particulate structure which changes according to environmental forcing (turbulence, light availability), particle dynamics, as well as organic and inorganic inputs and exports, whether they be autochthonous or allochthonous (Azam 1998, Azam and Malfatti 2007). All of these conditions feed back on one another resulting in a highly structured and dynamic environment where relative hotspots and coldspots exist with regard to microbial abundance and activity. Identification of conditions that create hotspot and coldspot locations will add much needed resolution to the operation and dynamics of the microbial loop and its effect on biogeochemical cycling of carbon and other important elements.

Blackburn et al. (1998) have found that phytoplankton cell lysis and excretion by protists create point source hotspots of microbial activity on the spatial and temporal scale of a few millimetres and 10 minutes, respectively. Seymour et al. (2000) found microscale heterogeneity in bacterial abundance on a 4.5 millimetre scale without the specific point sources that Blackburn et al. (1998) depended on. The temporal and spatial scales on which heterogeneity exists could also provide insight to the issue of bacterial growth efficiencies and the prevalence of active and inactive bacteria. Recent evidence suggests that microscale heterogeneity exists for bacterial metabolic activity in different aquatic bacterial communities (Seymour et al. 2004).

Generally, particulate organic matter (POM) is heavily colonized by bacteria and protists resulting in a hotspot of microbial activity compared to the surrounding water (Caron et al. 1986, Simon et al. 2002). This presumably occurs due to the relatively concentrated food source compared to bulk water, and colonization by organisms

specialized for use of solid substrates (Delong et al. 1993, Lemarchand et al. 2006). The concentration of microbes on particulate matter results in the efficient hydrolysis of the particulate due to increased efficiency of enzymatic hydrolysis compared to open water (Simon et al. 2002), although there seems to be exceptions (Tang et al. 2006). The result of this efficient hydrolysis is incomplete use of hydrolysed material, as shown by the net flux of labile DOM into the surrounding water and the associated DOM gradient (Smith et al. 1992, Kiorboe et al. 2001).

Accompanying these ideas of water column micro-structure is the discovery of new classes of particles such as carbon rich, transparent exopolymer particles (TEP) (Alldredge et al. 1993), protein containing, Coomassie stained particles (CSP) (Long and Azam 1996), and detritus rich, DAPI yellow particles (DYP) (Mostajir et al. 1995, Brachvogel et al. 2001, Carrias et al. 2002). The presence of these particles increases the numbers of previously disregarded, or at least undifferentiated, particles providing water column structure and potential hotspots for the microbial community. This is especially true of TEP due to their transparent nature. In the past, TEP colonized bacteria may have been enumerated as free-living bacteria, resulting in overestimates of that portion of the bacterial community and an underestimate of the particle associated community (Simon et al. 2002). It is also likely that different particle classes have different nutritional value for microbes.

The Mackenzie Delta contains a diverse set of lakes that exhibit different characteristics depending on their flooding frequency and duration. The spring flood is the major hydrologic event in the lakes, and ultimately acts as recharge for both water and nutrients. Differences in lake elevation dictate the frequency and duration of flooding. After the initial flood, in-lake processes begin to dominate and lake characteristics evolve over the open water season. The result is a variety of lakes in various stages of transition which exhibit a range of chemical and biological characteristics. This provides a convenient, natural ecosystem to test for differences in the bacterial response to particle structuring of the water column, as well as bacterial colonization of novel particles such as TEP.

During the open water season of 2006, we performed a simple budgeting exercise to assess the extent to which particles structure the water column for bacteria. Assessing the importance of TEP as a potential energy source and whether or not TEP represent potential hotspots for microbial activity was of special interest. We separated the free-living and attached bacterial communities suspended in the water column to gain insight into the microbial ecology of floodplain lakes. The basic model for the budget is:

Total bacteria = Free-Living bacteria + TEP-Attached bacteria + Other-Attached bacteria

where free-living bacteria are not associated with any particle type, TEP-attached bacteria include all bacteria associated with TEP and other-attached bacteria (OA-bacteria) represent what is not included in the previous two components. OA-bacteria would encompass bacteria attached to detritus or any non-TEP organic particle, bacteria attached to inorganic particles, and any TEP encased bacteria that could not be enumerated due to difficulties focussing through the particle.

We tracked changes in these bacterial components, in lakes over a range of sill elevations, to assess whether flooding frequency and duration affect budget components. We also tracked seasonal changes in the bacterial components. In addition to assessing bacterial colonization of TEP, we assess inter-lake and seasonal changes in TEP-attached bacterial densities. Concurrently, we measured in-situ lake conditions to probe for relationships explaining the observed trends.

4.3 Methods

Methodological information for this chapter has been described previously and will not be repeated here. The study area was described in section 3.3.1, and sample collection in 3.3.2. Detailed information on TEP slide preparation and microscopic TEP enumeration are located in section 2.3.3.1, and the methods for TEP-carbon calculation are described in section 2.3.4. Section 3.3.3 contains our methods for chlorophyll a (Chl a), total suspended solids (TSS), particulate organic carbon (POC), total dissolved organic carbon (TDOC), and coloured dissolved organic carbon (CDOC). The method used to quantify dissolved carbohydrates is located in section 3.3.4. Enumeration of TEP-attached, free-living, and total bacteria was done by the methods described in section 3.3.6. Please refer to in section 3.3.8 for details on data analysis.

4.4 Results

4.4.1 Study System Conditions

Chlorophyll a

Chlorophyll a concentrations ranged from 0.4 to 8.0 ± 1.7 μg/L over the course of the summer (Figure 4.1A). Lakes 56 and 520 reached peak Chl a concentration in the second week of sampling before algal populations declined over subsequent weeks. After the initial bloom, Chl a remained low in Lake 56 while Lake 520 exhibited a steady increase in Chl a from mid-July through August. Peak Chl a concentration in Lake 129 was reached during the 4^{th} week of sampling and declined rapidly thereafter before stabilizing for the remainder of the summer.

TDOC

TDOC concentrations ranged from 7.55 to 10.34 ± 0.73 mg/L (Figure 4.1B) with mean concentrations of 8.49, 9.79, and 8.43 mg/L in Lakes 129, 56, and 520 respectively. Lake 56 TDOC concentrations peaked as Chl a declined. There was a gradual decrease in TDOC in Lake 56 over the rest of the summer, although the actual concentrations were always higher than either Lake 520 or 129. TDOC concentrations began low in Lake 520 and steadily increased over the summer. Lake 129 had fluctuating TDOC concentrations but exhibited a general decrease over the summer and a sharp decline in August.

CDOC

CDOC began high and declined over the summer in all lakes (Figure 4.1C). Mean absorbencies at 330 nm were 0.439, 0.397, and 0.372 in lakes 29, 56, and 520. Therefore, CDOC concentrations increase with increasing flooding frequency.

Dissolved Carbohydrates

TDCHO ranged from 0.47 to 1.15 ± 0.14 mg-C/L (Figure 4.2A). Mean concentrations were very similar in all lakes (Lake 129, 0.80 mg-C/L; Lake 56, 0.86 mg-C/L; Lake 520, 0.81 mg-C/L). There was significant fluctuation in TDCHO from week to week but concentrations tended to decrease over the season in Lake 129 and 56 while concentrations increased in Lake 520.

Monosaccharide concentrations ranged from 0.31 to 0.83 ± 0.12 mg-C/L (Figure 4.2B) and mean concentrations were 0.57, 0.53, and 0.52 mg-C/L in Lakes 129, 56, and 520 respectively. Similarly to TDCHO, monosaccharide concentrations tended to decrease over time in Lake 129 and 56, and increased over time in Lake 520.

Polysaccharide concentrations ranged from 0.07 to 0.51 ± 0.09 mg-C/L (Figure 4.2C) with mean concentrations in Lakes 129, 56, and 520 being 0.23, 0.33, and 0.29 mg-C/L respectively. The seasonal trend was stable for Lake 129, and polysaccharide concentrations increased in both Lake 56 and 520.

TSS

Immediately after the flood, TSS concentrations are very high (up to 79.2 mg/L) and decline quickly as flood waters recede (Figure 4.3A). During the remainder of the summer TSS exhibit significant fluctuation, but mean concentrations are higher in lower elevation (high flood frequency) lakes if the initial sampling date is not considered (Lake 129, 7.4 mg/L; Lake 56, 6.9 mg/L; Lake 520, 4.0 mg/L).

POC

POC follows a similar trend as TSS (Figure 4.3B). All lakes exhibit a steady decline from initially high post-flood concentrations. Mean POC concentrations were 1.08, 1.23, and 0.84 mg/L in lakes 129, 56, and 520 respectively.

4.4.2 Bacteria Budget

Total Bacteria

Total bacteria abundances ranged from 30.2 x 10^5 to 167.5 x 10^5 cells/ml with a mean of 82.6 x 10^5 ± 31.1 x 10^5 cells/ml in all lakes surveyed. In Lake 129, total bacteria numbers were initially high and stabilized near the mean as the summer proceeded. There is no clear pattern for total bacteria abundances in Lake 56 and abundances in Lake 520 exhibited a steady decline over the summer. These seasonal trends are shown in Figure 4.4 and descriptive statistics are in Table 4.1. The results of the randomized block ANOVA (Table 4.2) found insignificant time effects ($F = 2.20$, $p = 0.086$, $df = 8$) and lake effects ($F = 3.16$, $p = 0.070$, $df = 2$) on total bacterial abundances.

TEP-Attached Bacteria

Mean TEP-attached bacteria abundances ranged from 1.57 x 10^5 to 14.97 x 10^5 cells/ml and had the lowest mean abundances (7.25 x 10^5 ± 3.60 x 10^5 cells/ml) of all bacterial fractions, in all lakes surveyed (Table 4.1). There were no clear seasonal trends in either Lake 129, 56, or 520 and there was no significant time or lake effect for TEP-attached bacteria abundance in the lakes surveyed (Table 4.2). Interestingly, each lake reached their maximum and minimum TEP-attached bacteria abundances at different times of the season (Figure 4.4), and didn't necessarily track TEP abundance very closely. This is evident from the poor relationship between attached bacteria and TEP (r^2 = 0.01) or TEP_{carbon} (r^2 = 0.12) in Lake 56 (Table 4.5).

Free-Living Bacteria

Free-living bacteria abundances ranged from 2.52 to 27.52 x 10^5 cells/ml with a mean of 10.50 x 10^5 ± 6.29 x 10^5 cells/ml. In Lake 129 and 520, free-living bacteria abundances were characterized by a gradual seasonal decrease, punctuated by midsummer spikes in abundances. Free-living bacteria abundances fluctuated greatly from week to week in Lake 56 (Figure 4.4). The randomized block ANOVA (Table 4.2) found a marginally insignificant time effect (F = 2.56, p = 0.056, df = 8) and a significant lake effect (F = 5.58, p = 0.015, df = 2).

Other-Attached Bacteria

In Lake 129 and 520, OA-bacteria abundances started high and decreased towards the end of the summer while abundances in Lake 56 fluctuated around the mean (Figure 4.4). There was a significant time effect (F = 3.17, p = 0.026, df = 2) and a marginally insignificant lake effect (F = 3.42, p = 0.060, df = 8). The seasonal pattern for OA-bacteria is very similar to that of total bacteria.

Expressing the bacterial fractions as a percentage of total abundances, OA-bacteria constitute > 75% of the total bacteria in the lakes surveyed (Figure 4.5 and Table 4.1). TEP-attached bacteria almost always contributed the smallest fraction of total bacteria (Mean 9.55%), while free-living bacteria contributed a mean of 13.91% of total

bacteria in all lakes combined. The component abundances, as a percent of total bacteria, are shown for each lake and sampling date in Figure 4.5.

Correlation with Bacterial Abundances

We assessed the relationship between a suite of relevant variables and bacterial abundances from each component of the bacteria budget, for each lake individually, as well as with all lakes combined. Relationships are assessed according to the coefficient of determination and the slope direction (positive or negative). The results are presented in Table 4.5 and we will highlight significant relationships here.

When all lakes were combined, total bacteria had a significant, positive relationship with TEP, TEP_{carbon}, TSS, and CDOC. Interestingly, there is no correlation between Chl a and bacteria abundance. In Lake 129, significant negative relationships were found between total bacteria and polysaccharides and carbohydrates. In Lake 520, there were significant positive relationships between total bacteria and CDOC, POC, TEP_{carbon}, and TEP while significant negative relationships were found for TDOC, polysaccharides, and Chl a.

TEP-attached bacteria had significant positive relationships with TEP and TEP_{carbon} when all lakes were combined. Interestingly, there was always a better relationship between TEP_{carbon} and TEP-attached bacteria than TEP volume concentration. There was also a significant relationship between TEP-attached bacteria and CDOC in Lake 520.

When all lakes were combined there was a significant positive relationship between free-living bacteria and TDOC. However, in Lake 520, there was a strong, negative relationship between free-living bacteria and both TDOC and TDCHO.

OA-bacteria was significantly correlated with TEP, TEP_{carbon}, CDOC, and TSS when all lakes were combined. Significant, negative relationships between OA-bacteria and both TDCHO and polysaccharide concentrations were found in Lake 129. In Lake 520, there were significant, positive relationships between CDOC, POC, TEP, TEP_{carbon}, and OA-bacteria while there were significant, negative relationships between OA-bacteria and both Chl a and polysaccharide concentration.

4.4.3 TEP-Attached Bacterial Colonization Density

Mean TEP-attached bacterial densities ranged from 0.220 – 0.387 cells μm^{-2} in the smallest size interval (Mean ESD = 3 μm), and 0.054-0.082 cells μm^{-2} in the largest size interval (Mean ESD = 48 μm) (Figure 4.6 and Table 4.3). We ran an ANOVA to assess whether there was a time effect or a lake effect on TEP-attached bacterial densities at each size class, as defined by mean ESD (Table 4.4). There was a significant lake effect in all size classes and a time effect on the intermediate size classes, 12 and 24 μm ESD. The TEP-attached bacterial density results show that the higher elevation lakes (56 and 520) have higher mean TEP-attached bacterial densities, especially for smaller particles (Table 4.3 and Figure 4.6).

TEP-attached bacterial densities obtained for the first sampling date that counts were possible (June 17), were equal among all lakes (Figure 4.7). This was also the date that the lowest TEP-attached bacterial densities were found. TEP-attached bacterial densities quickly diverge after this first sampling date. An example of this for a single size class is shown in figure 4.7. The general trend is increasing TEP-attached bacterial density from the beginning of the summer toward the end of summer (Figure 4.7).

Correlation with TEP-attached bacterial densities

We found correlation between with TEP-attached bacterial density and a number of variables. These relationships may provide insight to why densities differ among lakes. The data were analysed by aggregating the results from all lakes as well as examining the trends in individual lakes using linear regression. The results are compiled in Table 4.6.

We found that CDOC, POC, and Chl a generally had inverse relationships with TEP-attached bacterial densities, and were significant for some size intervals in Lakes 129 and 56. TSS also had a general negative relationship with TEP-attached bacterial densities, but was only significant in the frequently flooded Lake 129. TEP volume concentration and TEP_{carbon} also show a general negative trend and exhibit a significantly negative relationship with TEP-attached bacterial densities for some size intervals in Lakes 129 and 56.

Other significant results include a negative relationship with TEP-attached bacterial density for TDCHO and polysaccharide concentration in Lake 129. However, a positive relationship was found for TDCHO in Lake 520 and a positive relationship for polysaccharide concentration in Lake 56. There was also a significant, inverse relationship between TEP-attached bacterial density and TDOC in Lake 56. There was a significant positive relationship between monosaccharide and TEP-attached bacterial density for one size interval in Lake 520.

4.5 Discussion

4.5.1 Bacteria Budget

Total Bacteria

Our total bacterial abundances are higher than what has generally been reported for arctic lake ecosystems. Studies of Alaskan, Toolik Lake have consistently yielded bacterial abundances in the range of 1-20 x 10^5 cells/ml (Hobbie et al. 1983, Obrien et al. 1992), which is similar to abundances found in other arctic lakes (Bertilsson et al. 2003). Previous studies in the Mackenzie Delta have obtained low (Spears and Lesack 2006) and similar (Febria 2005) estimates in comparison to these other arctic lake ecosystems as a pre-filtration step was used in order to focus on free-living bacterial populations. Our higher estimates of bacterial abundance compared to previous Mackenzie Delta studies can be attributed to differences in methods, and evolving techniques which seem to be refining our ability to estimate total bacterial abundances, especially when suspended sediment concentrations are high. Our total bacterial abundances are at times similar to Febria (2005), but our maximum abundances are within the range found in hypertrophic systems (Sommaruga and Robarts 1997). We feel that our methodological refinements account for the observed, high, total bacteria abundances through enumerating a greater number of particulate attached bacteria via the use of a surfactant (TWEEN 80) plus sonication (Yoon and Rosson 1990). At the very least, our values are an appropriate index for total bacteria in this system.

Planktonic bacterial production and abundances increase with planktonic primary production and Chl a across a wide range of aquatic ecosystems (Cole et al. 1988). However, this was not the case in the lakes we surveyed in the Mackenzie Delta. Exceptions to the pattern include ecosystems where there are significant contributions from alternate carbon sources which include macrophytes, suspended sediment re-suspension, and allochthonous inputs (Findlay et al. 1991). The presence of differing amounts of these alternate carbon sources in the lakes surveyed here, could account for the lack of relationship between bacterial abundances and Chl a, and the inverse relationship found in Lake 520 (Table 4.5). When the lakes are combined, total bacteria

abundances seem to be positively influenced by an allochthonous carbon signal. This is evident in the relationship between total bacteria and both TSS and CDOC which are strongly related to river water inputs and terrestrial carbon contributions. The insignificant relationship between POC and total bacteria in all lakes combined is surprising, however POC seems to be of greater importance in Lake 520.

TEP-Attached Bacteria

Comparisons of TEP-attached bacteria abundances are best done by looking at abundances relative to total bacteria. We found that mean attached bacterial abundances were ~10% of total bacterial abundances in all the lakes surveyed, although there was some deviation throughout the season (Figure 4.5). This level of TEP colonization is typical if not slightly higher than values found in both marine (Passow 2002a and references therein) and freshwater systems (Worm and Sondergaard 1998a, Carrias et al. 2002).

Our results support the hypothesis that bacterial colonization of TEP is more closely related to the amount of substrate (in this case carbon) rather than volume concentration. This assumes that the empirical equation used to obtain our TEP_{carbon} estimates is appropriate for TEP formed in Mackenzie Delta Lakes. However, it is clear that there are other major factors affecting bacterial colonization of TEP.

Free-Living Bacteria

Our free-living bacteria abundance estimates should be comparable to the bacteria abundance estimates of Spears and Lesack (2006) as they filtered out any particulates using a GF/C filter ($0.39 - 0.82 \times 10^{-5}$). However, we yielded an order of magnitude more free-living bacteria than they did (Table 4.1). We cannot account for these differences, but it seems that the choice of methods, coupled with large apparent spatial and temporal variability results in a wide range of bacterial abundance estimates in Mackenzie Delta Lakes (Teichreb 1999, Febria 2005, Spears and Lesack 2006). The positive relationship between TDOC and free-living bacteria abundances highlights the general importance of DOC as a substrate for free-living bacteria. However, when analysed separately there were no significant relationships in either Lake 129 or 56 and a negative relationship in

Lake 520 between free-living bacteria and both TDOC and TDCHO. This is perplexing as TDCHO should represent a relatively labile carbon source for free-living bacteria.

Other-Attached Bacteria

Since OA-bacteria, as described here, are likely dominated by bacteria attached to non-TEP particulate matter, we see a general decrease in bacteria numbers as the summer proceeds and TSS decrease. We also observe a river water signal shown by the positive relationship between OA-bacteria and both CDOC and TSS, which suggests the importance of allochthonous carbon sources. However, looking at the relationships between TEP, TEP_{carbon}, and OA-bacteria abundance, there is a strong possibility that a large portion of OA-bacteria may actually be TEP encased. TEP encased bacteria can be difficult to enumerate due to difficulties focusing through TEP particles and variability in the transparency of the particle. We cannot confirm that TEP encased bacteria contribute greatly to the OA-bacteria numbers, but this possibility should be examined as the proportion of TEP-attached bacteria may be much greater than we have estimated here.

By adding TEP-attached bacteria to OA-bacteria, we can estimate total attached bacteria. Doing this gives a mean total attached bacteria abundance of 86% of total bacteria. In other ecosystems, the relative proportion of attached bacteria is variable, but levels this high are generally associated with ecosystems exposed to high levels of hydrodynamic forcing, such as rivers and estuaries, which have proportions ranging from 14 – 90% (Zimmermann-Timm 2002). Lacustrine ecosystems generally have lower relative proportions of attached bacteria (5 - 30%; Grossart et al. 1998), although there are exceptions for shallow lakes (> 50%; Wu et al. 2007), or up to 80% in an enclosure experiment (Worm et al. 2001). These studies likely underestimate attached bacteria by enumerating TEP-attached bacteria as free-living, however, this omission would likely cause a difference of no more than 5-10%. Our attached bacteria numbers are at the high end of those reported for lake ecosystems, and highlight the importance of suspended particulate matter for bacteria in Mackenzie Delta Lakes. It also suggests that the microbial loop plays an important role in the turnover of particulate carbon and recycling of nutrients from POM. The proportion of metabolically active cells, among bacterial

fractions within lakes of different flood frequency and duration, should be a priority for future research in the Mackenzie Delta.

4.5.2 TEP-Attached Bacterial Colonization Density

It has been well established that while larger TEP generally have more bacteria attached to them, smaller TEP are more densely colonized by bacteria (Passow and Alldredge 1994, Mari and Kiorboe 1996, Carrias et al. 2002). Our results confirm that this is true for TEP in lakes of the Mackenzie Delta. Our estimates of mean TEP-attached bacterial density are within the range reported for marine ecosystems (Passow 2002a and references therein) and are remarkably similar to those reported by Carrias et al (2002) for an oligomesotrophic and eutrophic lake.

We found that mean TEP-attached bacterial densities were significantly higher, in higher elevation lakes (lower flooding frequency). If bacterial density is taken as an indication of substrate importance, our results suggest that TEP formed in higher elevation lakes is, on average, more important for the bacterial community than TEP formed in lower elevation lakes. Other authors have suggested that differences in TEP-attached bacterial densities are due to differences in TEP particle age, TEP particle density (Schuster and Herndl 1995), or fractal dimension (Mari and Kiorboe 1996). Therefore, differences in bacterial colonization of TEP could be related to differences in TEP formation and removal, the source of TEP precursors, or grazing pressures depending on lake elevation. Considering the differences in connection times (allochthonous inputs), size (wind mixing), and food web structure between the lakes, it is difficult to identify the main factor controlling TEP-attached bacterial densities.

TEP-attached bacterial densities were equal in all 3 lakes immediately after the spring flood (June 17), suggesting that TEP was of equal bacterial importance in all lakes at this time. TEP enumerated on this sampling date may still be largely of a riverine origin in all lakes as it was not long after the flood. Therefore, on this date, TEP would have a common source and formation mechanism. The lowest TEP-attached bacterial colonization density for almost all lakes and size classes found during the 2006 season occurred at this time and could be an indication that riverine derived TEP is of low

quality (or at least low importance) compared to the TEP enumerated later in the summer. Another interpretation would be that bacteria would not colonize TEP particles due to the higher abundance of other organic particulates, and higher concentrations of dissolved nutrients that are associated with flood waters (Lesack et al. 1998). A third explanation invokes TEP turnover time. TEP may be rapidly formed and removed via sedimentation at this time of year due to the high levels of suspended sediments, resulting in limited colonization of all particle sizes. However, it appears that particles are colonized by bacteria on the order of seconds to minutes (Kiorboe et al. 2002) so rapid TEP turnover is not likely limiting bacterial colonization.

As the summer proceeds, mean TEP-attached bacterial densities increase. Although the increase was not statistically significant in all size classes, we feel that this is an important result and could indicate a shift in the microbial communities in Mackenzie Delta Lakes. The decrease or absence of riverine inputs later in the summer means that autochthonous sources likely contribute a greater proportion of TEP-precursors to TEP formation than immediately after the flood. Therefore, TEP formed from autochthonous production is of greater importance for bacteria in Mackenzie Delta Lakes compared to TEP formed from allochthonous sources. This could imply that TEP formed from these autochthonous sources is more labile than TEP formed in the early summer. Alternately, it could mean that TEP represent a more favourable substrate during this time due to decreases in particulate matter, dissolved nutrients, and the potential for TEP to scavenge colloids and amino acids (Decho 1990).

There is some evidence to support the hypothesis that organic particles compete with TEP for bacterial colonization. The regression analysis of TEP-attached bacterial densities versus a suite of variables found that densities were generally lower when organic particulates, phytoplankton (Chl a), POC, and TEP were at a higher concentration. Indeed, it appears that TEP itself provides intraspecific competition by increasing the number of colonization sites. CDOC also has a general and often significant negative relationship with TEP-attached bacterial densities. Since CDOC is a strong signal of river water influence, the initial input of river water, which carries with it high concentrations of detritus as well as nutrients, could decrease the importance of TEP for bacterial use. POC and phytoplankton may be important substrates for bacteria in

these lakes and likely represent a higher food quality than carbon enriched TEP. The presence of a general, but insignificant negative relationship between TEP-attached bacterial densities and TSS suggests that inorganic materials are not competing substrates for bacterial colonization.

The potential for differences in substrate quality within particles of the same class (TEP) highlights the heterogeneous nature of particles formed from the colloidal fraction of DOC, and the heterogeneous nature of DOC itself. Adding to this complexity is the potential for the inclusion of small amounts of detritus in TEP which could greatly alter its nutritional value. We cannot comment directly on the quality of organic matter that composes TEP in different lakes, however, we can discuss the DOC dynamics in Mackenzie Delta Lakes which may have an impact on TEP precursors and TEP quality.

The dynamics of DOC in Mackenzie Delta Lakes have been relatively well defined and depend on sub catchment runoff, river water inputs (sill elevation), and macrophyte biomass (Lesack et al, unpublished). Sub-catchment runoff may add a significant amount of DOC to lakes but due to low average precipitation levels, this effect is variable and generally constrained to a similar time frame as the spring flood (Marsh 1986). In low elevation lakes, TDOC concentrations generally track river concentrations (Lesack et al, unpublished). The presence of high macrophyte biomass tends to increase the TDOC concentration of lakes due to leaching and decomposition (Squires and Lesack 2003a). Therefore, once a high-sill lake becomes detached from the river channel, TDOC concentrations generally increase if macrophyte biomass is high. This results in a general trend where TDOC increases as flood frequency decreases. Conversely, CDOC concentrations decline over the summer due to photobleaching (Gareis 2007), and generally increase with increasing flood frequency.

Our study showed the characteristic seasonal CDOC decline, and mean CDOC concentrations increased with increasing flood frequency. Our TDOC results show declining concentrations in, low elevation, Lake 129 which has a moderate macrophyte biomass (*Potamogeton spp.*). There was little net change in, high elevation, Lake 56 which has a high macrophyte biomass (*Potamogeton spp.*). A steady increase in TDOC was observed in, high elevation, Lake 520 which has a high macrophyte biomass (*Chara*

spp. and Ceratophyllum spp.). Although our data do not follow, perfectly, the pattern of DOC dynamics described above, we can at least infer that higher macrophyte biomass buffers TDOC declines and can cause TDOC to increase. Both algal and angiosperm macrophyte derived DOC has been shown to aggregate (Alber and Valiela 1994, Thornton 2004), and therefore, TEP in higher elevation lakes, with high macrophyte biomass, could have a different TEP precursor source than lower elevation lakes. This could increase the suitability of TEP as a substrate resulting in the higher bacterial densities we have observed in these high elevation lakes. There is also a significant epiphyton biomass colonizing macrophytes in the lakes. It is unknown what their contribution to DOC and TEP-precursors are, but it could be significant.

It is well established that protistan grazing has a significant effect on planktonic bacterial abundance in aquatic ecosystems, and can account for the removal 0-200% of bacterial production (Sigee 2005). Heterotrophic flagellates have been identified as major contributors to these high grazing rates, and their populations can double on the order of days (Berninger et al. 1991). In some cases small ciliates also exert high grazing pressure on bacterial populations as well (Simek and Straskrabova 1992). There are few studies that include protistan grazing on attached bacteria, but emerging evidence is showing that attached bacteria are also subjected to high grazing pressure and particulates represent an important micro-niche for certain protists (Artolozaga et al. 2000, Artolozaga et al. 2002, Kiorboe et al. 2003, Kiorboe et al. 2004, Tang et al. 2006). We believe that a sufficient answer for patterns in bacterial abundances and densities will not be possible without estimating heterotrophic protistan abundances and grazing rates. Identification of the importance of protistan grazing, and the control of bacterial populations, is also important for estimates of microbial loop contributions to the classical food web. These contributions could be significant due to the abundance of organic carbon and prevalence of nutrient limitation in Mackenzie Delta Lakes. Heterotrophic flagellates and bacteria were concurrently enumerated in some Mackenzie Delta Lakes by Riedel (2002) during the summer of 2001. However, abundances of bacteria and heterotrophic flagellates were found to be very low compared to the bacterial abundances reported here. Riedel (2002) also found complex trophic cascades affected abundances in grazing experiments.

The influence of grazing activity on attached bacteria abundance and density distribution is unknown for Mackenzie Delta Lakes. Given the propensity for heterotrophic protists to control both free-living and attached bacteria in a variety of aquatic ecosystems (Berninger et al. 1991, Kiorboe et al. 2004, Tang et al. 2006), this is an important avenue for future research. Heterotrophic protists may provide an important link for carbon and nutrient flow between secondary bacterial production and the classical food web, sustaining a higher biomass of zooplankton and fish than can be explained by autotrophic sources alone.

4.6 Conclusion

Through a simple budgeting exercise, we have quantified the proportion of the bacterial community that are free-living in the water column or attached to particulate matter in Mackenzie Delta Lakes. We enumerated total, free-living, and TEP-attached bacteria to get an estimate of the contribution of free-living vs. TEP-attached vs. other-attached bacteria to the community of total suspended bacteria. Total bacteria abundances were significantly higher immediately after the spring flood and decreased towards the end of August. Results show that ~5-10% of bacteria are attached to TEP, at any time, among 3 lakes of differing flooding frequency and duration. Strong correlation between TEP and OA-bacteria suggest that a significant number of bacteria may actually be TEP-encased, resulting in an underestimate of TEP-attached bacteria. A surprisingly low proportion of bacteria in the lakes surveyed were actually free-living (~10-20%). Results here indicate an average of ~85% of total suspended bacteria are attached to some type of particle in Mackenzie Delta Lakes. This is among the highest proportions of particulate associated bacteria thus far reported in any aquatic ecosystem, and highlights the importance of particulate substrates for the planktonic bacterial community in floodplain lakes.

As reported by other authors (Passow and Alldredge 1994, Mari and Kiorboe 1996, Carrias et al. 2002), TEP-attached bacterial densities characteristically decline as TEP size increases. TEP-attached bacterial densities increased from post-flood conditions to late-summer, although the result was only significant in intermediate size classes. Additionally, TEP-attached bacterial densities were higher in lakes that flood less frequently and for a shorter duration (higher elevation). Strong negative correlation with particulate organic matter suggests that the number of favourable attachment sites play a role in regulating inter-lake and seasonal changes in TEP-attached bacterial densities.

4.7 Figures

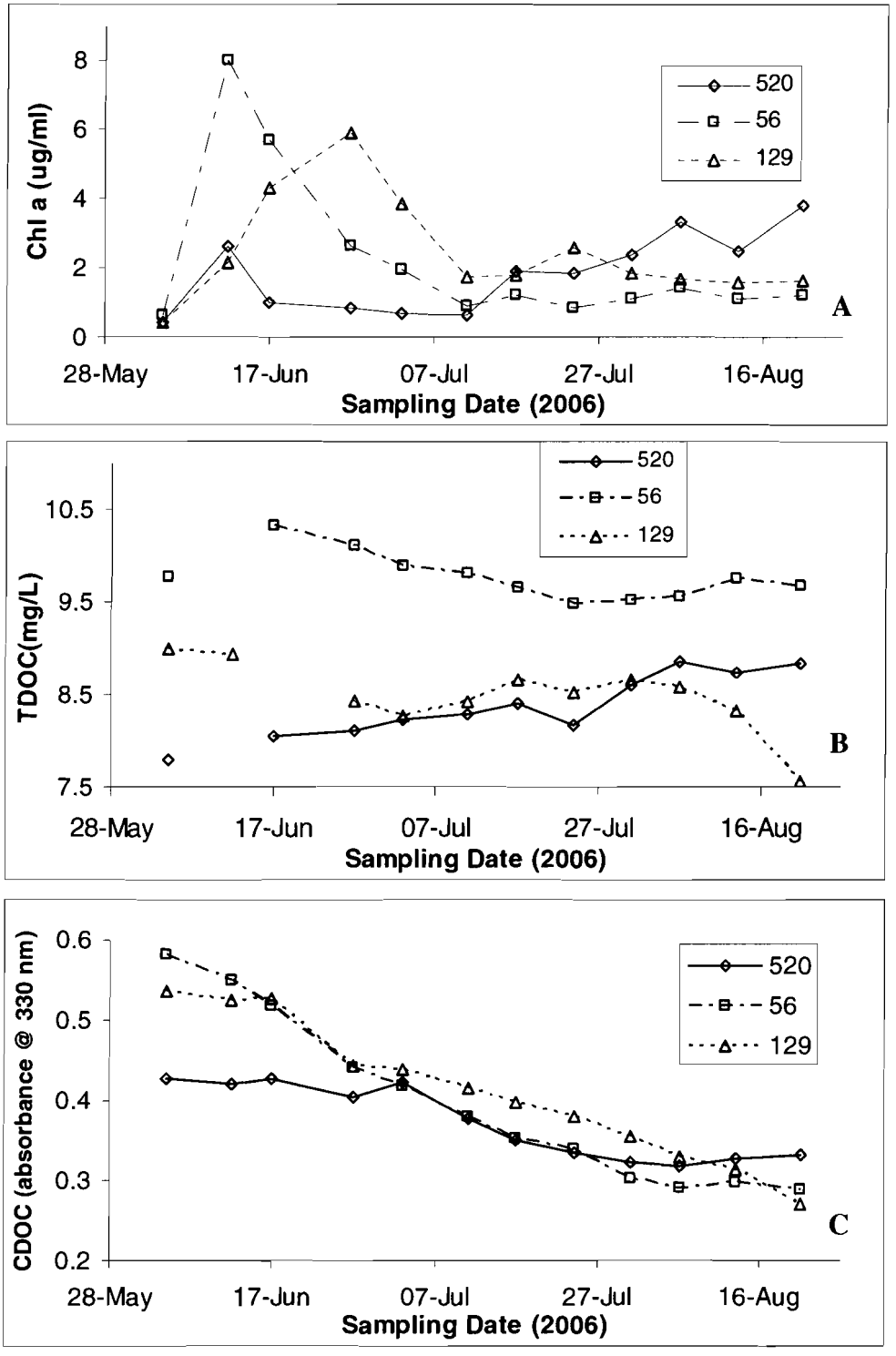
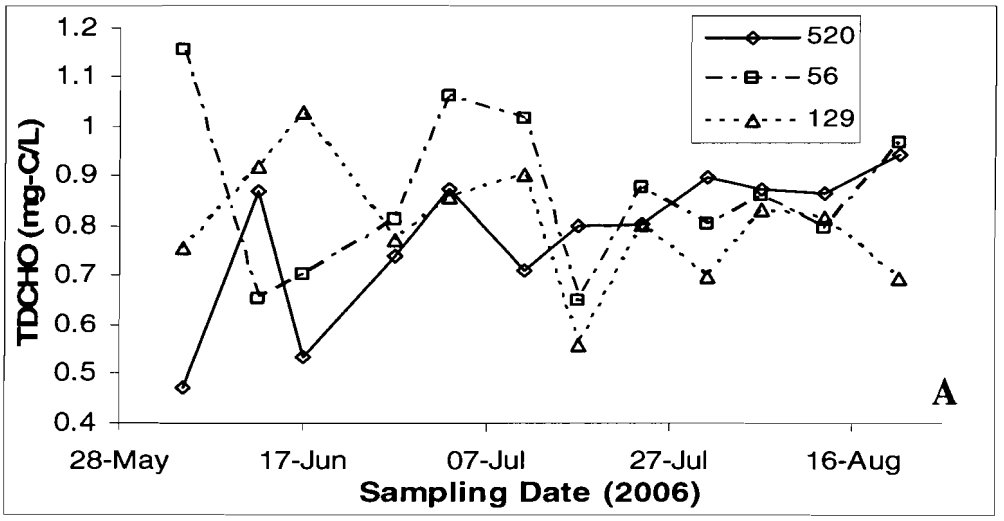
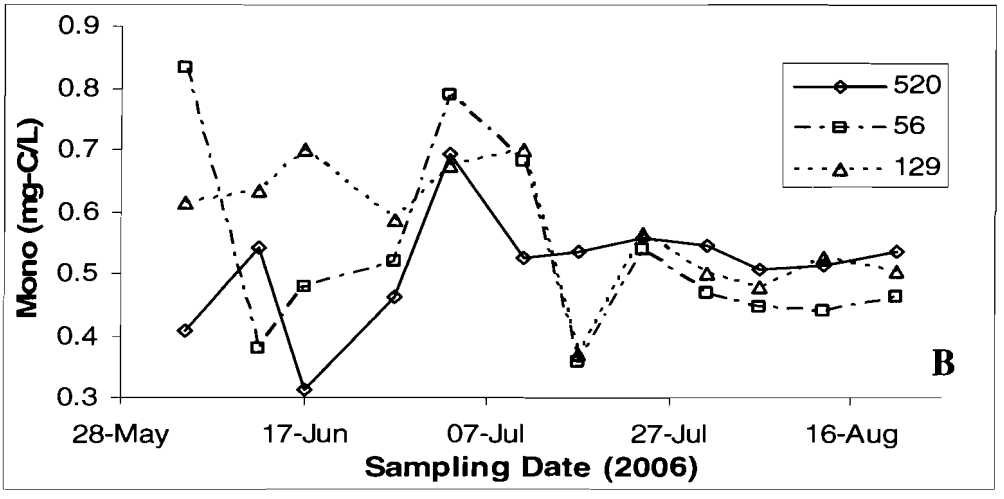


Figure 4.1 **Seasonal trends for A) Chl a, B) TDOC, and C) CDOC for Lakes 520, 56, and 129 during the open water period in 2006.**





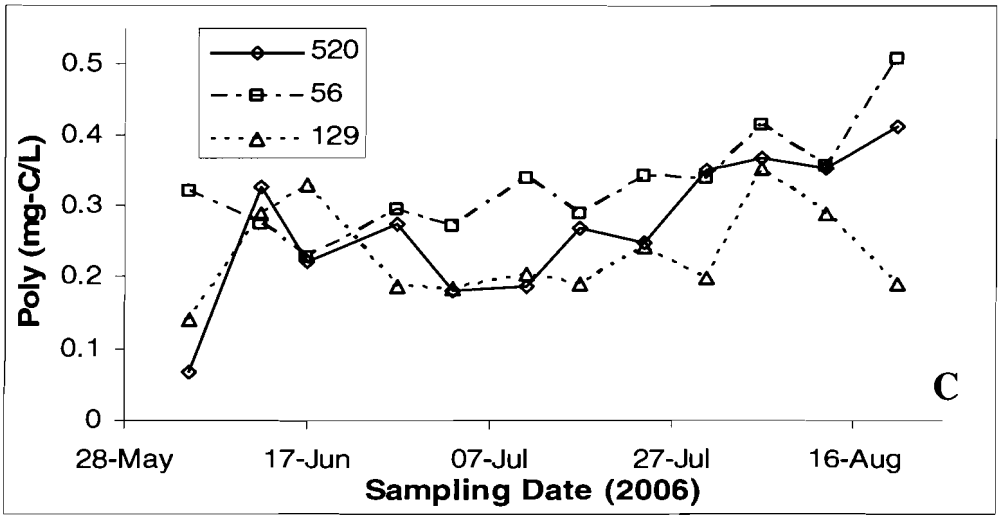


Figure 4.2 **Seasonal trends for carbohydrates in Lakes 520, 56, and 129 during the open water period in 2006. A) TDCHO, B) Monosaccharide , and C) Polysaccharide concentrations.**

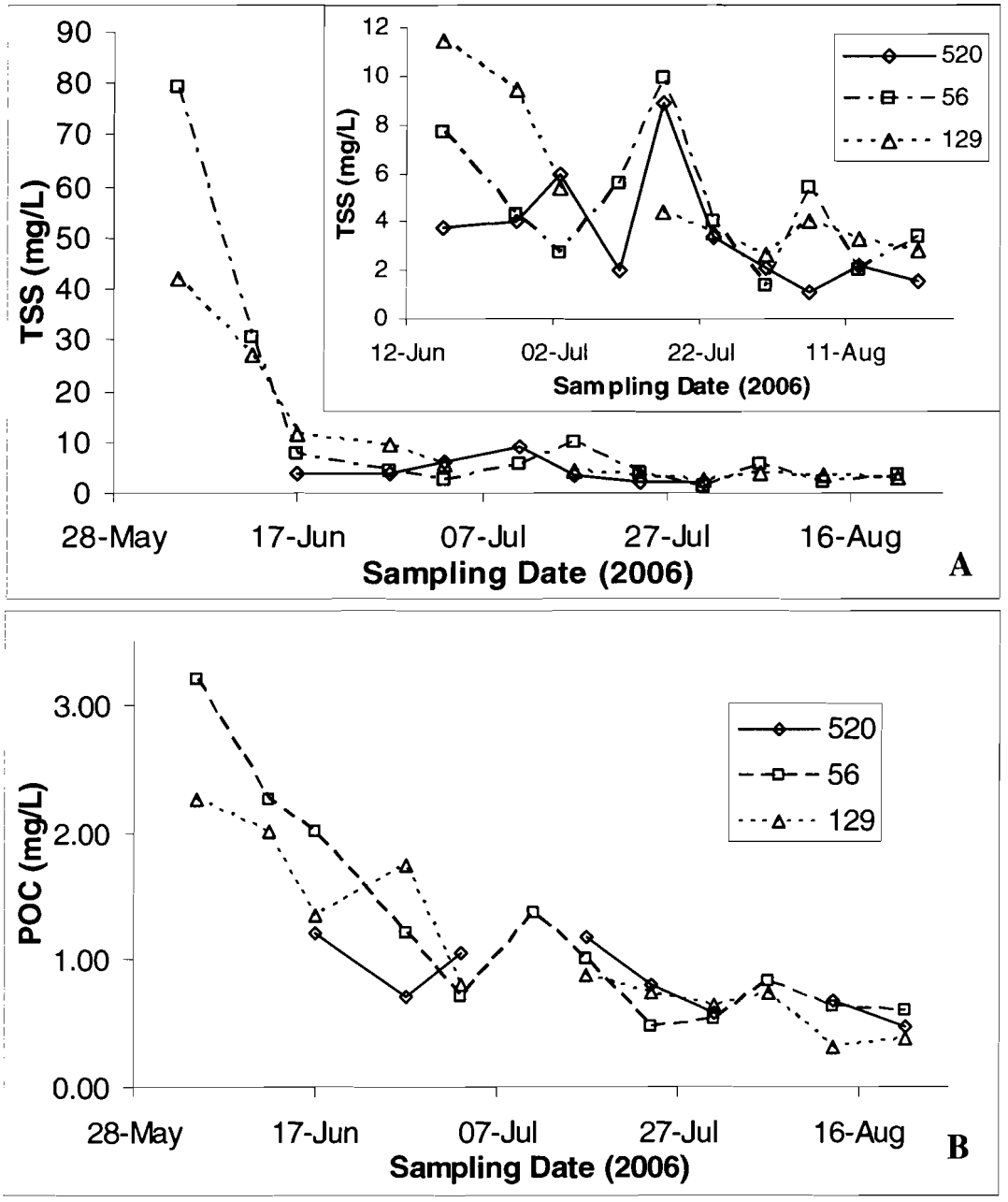


Figure 4.3 Seasonal trends in A) TSS (plus inset showing greater detail) and B) POC for Lakes 520, 56, and 129 over the 2006 open water season.

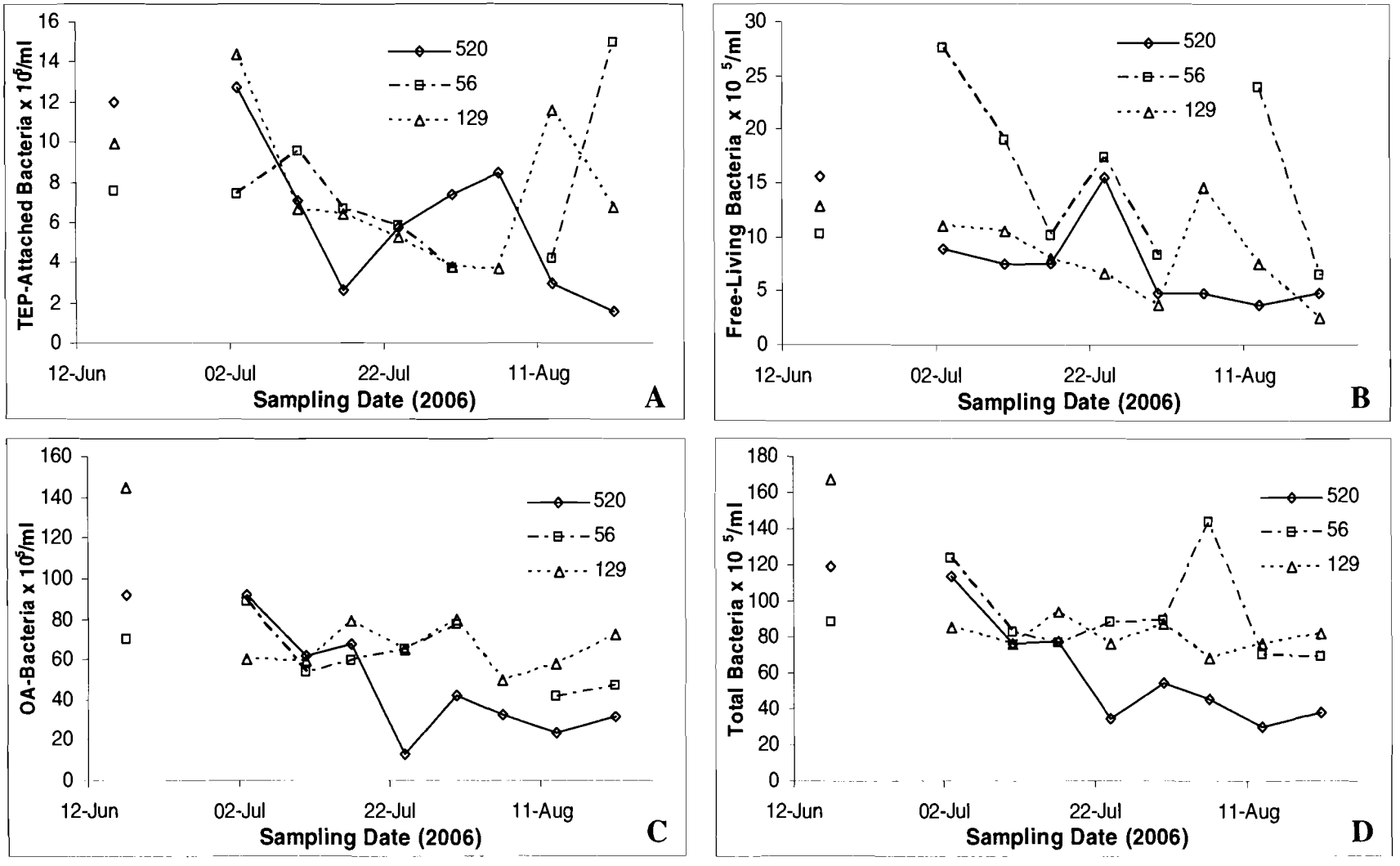


Figure 4.4 **Seasonal bacteria abundances for A) TEP-attached, B) free-living, C) other attached- (OA-), and D) total bacteria in Lake 520, 56, and 129.**

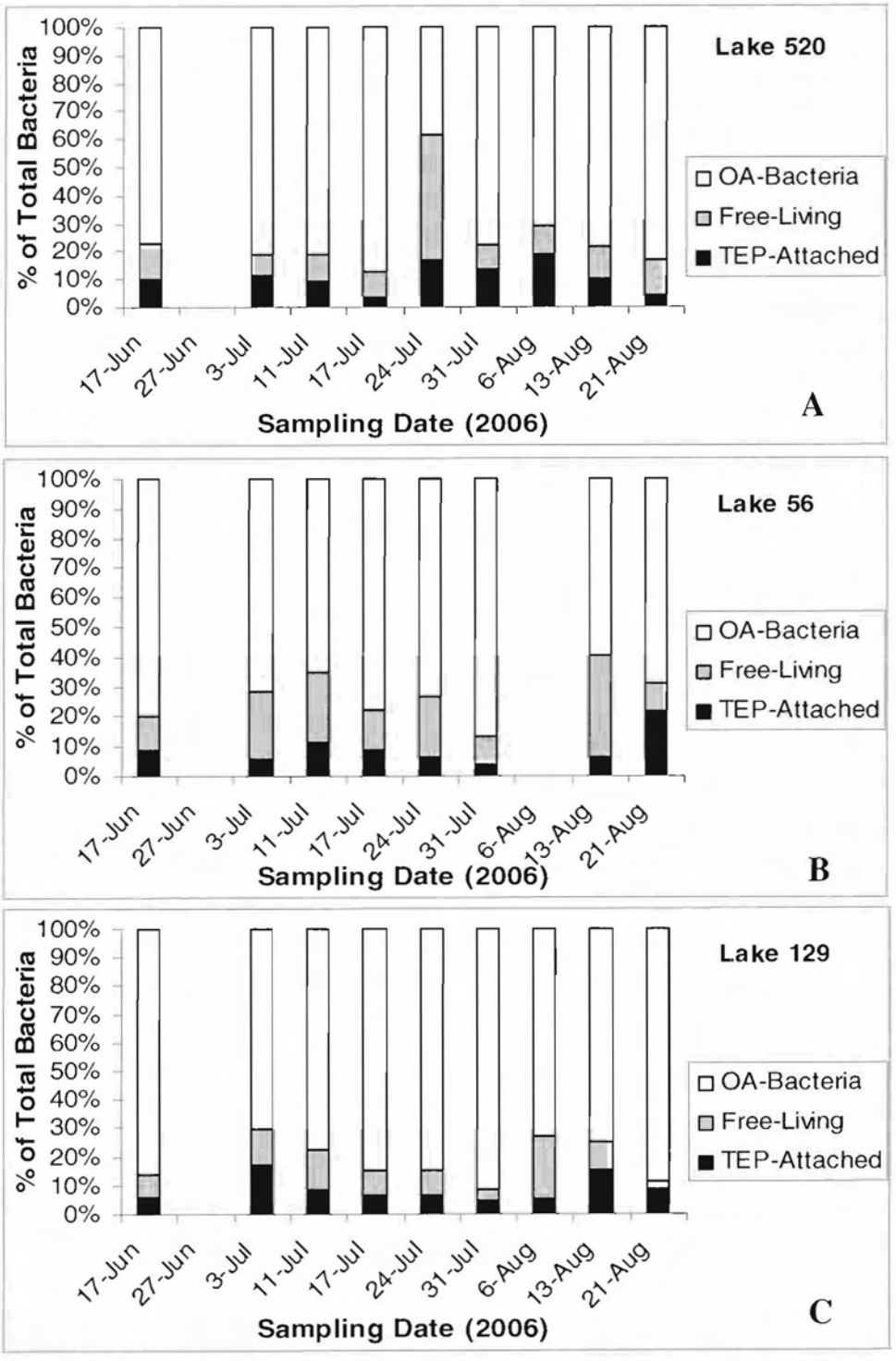


Figure 4.5 **Proportion of total bacteria contributed by TEP-attached, free-living, and OA-bacteria in Lakes A) 520, B) 56, and C) 129 during the summer of 2006.**

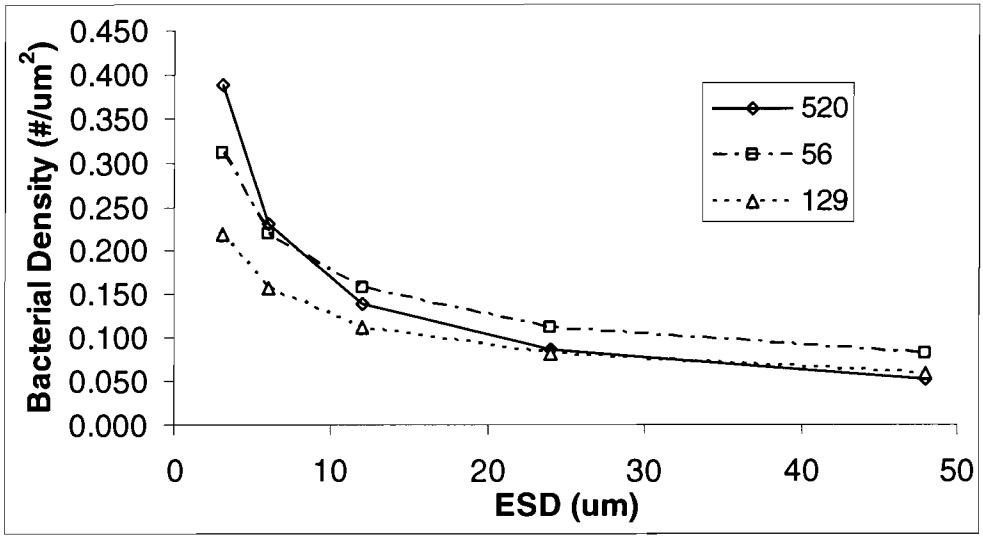


Figure 4.6 Mean TEP-attached bacterial densities (# cells/μm^2) for each size class (mean ESD) for lakes 520, 56, and 129.

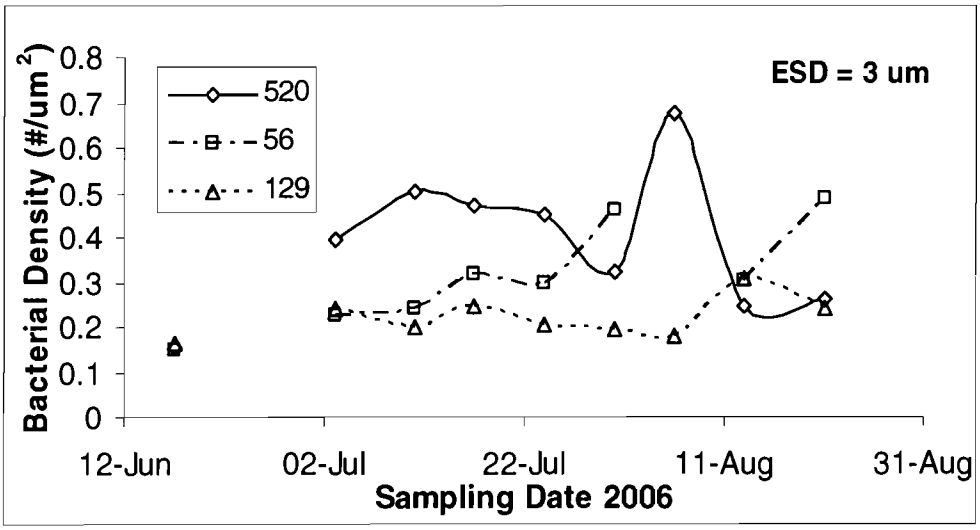


Figure 4.7 A sample plot showing the divergence of TEP-attached bacterial densities (# cells/μm^2) of the same size class (Mean ESD = 3 μm) from an equal value at the beginning of the season in lakes 520, 56, and 129.

4.8 Tables

Table 4.1 **Descriptive statistics for bacterial abundances in each component for lakes 129, 56, 520, and all lakes combined. Values are bacterial abundances (# cells x 10^5/ml).**

		N	Mean	SD	Min	Max	Mean % of Total Bacteria
Lake 129	TEP-Attached Bacteria	9	7.59	3.63	3.69	14.39	8.70
	Free-living Bacteria	9	8.59	4.06	2.52	14.55	9.99
	Other-Attached Bacteria	9	74.16	28.32	49.93	144.73	81.31
	Total Bacteria	9	90.34	29.88	68.17	167.55	-
Lake 56	TEP-Attached Bacteria	9	6.73	3.97	1.57	12.74	9.16
	Free-living Bacteria	9	8.09	4.53	3.64	15.62	17.87
	Other-Attached Bacteria	9	50.68	28.89	13.38	91.97	72.97
	Total Bacteria	9	65.50	33.43	30.23	119.22	-
Lake 520	TEP-Attached Bacteria	8	7.46	3.57	3.65	14.97	10.75
	Free-living Bacteria	8	15.35	7.76	6.38	27.52	14.31
	Other-Attached Bacteria	8	62.83	15.60	41.49	88.59	74.94
	Total Bacteria	9	92.05	25.14	68.71	143.34	-
All Lakes	TEP-Attached Bacteria	26	7.25	3.60	1.57	14.97	9.55
	Free-living Bacteria	26	10.50	6.29	2.52	27.52	13.91
	Other-Attached Bacteria	26	62.54	26.29	13.38	144.73	76.54
	Total Bacteria	27	82.63	31.08	30.23	167.55	-

Table 4.2 **Results of the randomized block design ANOVA with Lake as the random effect and time as the fixed effect.**

	N	df	F-Stat	p-value
Time effect				
Total Bacteria	27	8	2.1967	0.0858
Free-living Bacteria (log transformed)	26	8	2.5584	0.0556
TEP-Attached Bacteria	26	8	1.0835	0.4244
Other-Attached Bacteria	26	8	3.1711	0.0258
Lake effect				
Total Bacteria	27	2	3.1555	0.0700
Free-living Bacteria (log transformed)	26	2	5.5788	0.0154
TEP-Attached Bacteria	26	2	0.1276	0.8811
Other-Attached Bacteria	26	2	3.4230	0.0596

Table 4.3 **TEP-attached bacterial densities (# cells/μm^2) calculated for each sampling date and size interval (Mean ESD). The mean TEP-attached bacterial densities for the entire 2006 season are included. We were unable to determine bacterial densities for some sampling dates as indicated by 'nd'.**

	Mean ESD (μm)	17-Jun	27-Jun	3-Jul	11-Jul	17-Jul	24-Jul	31-Jul	6-Aug	13-Aug	21-Aug	Mean
Lake 129	3	0.166	nd	0.243	0.202	0.248	0.203	0.193	0.178	0.307	0.241	0.220
	6	0.110	nd	0.160	0.160	0.180	0.145	0.148	0.134	0.188	0.186	0.157
	12	0.073	nd	0.106	0.127	0.131	0.103	0.114	0.101	0.115	0.143	0.113
	24	0.049	nd	0.070	0.101	0.095	0.074	0.088	0.076	0.070	0.111	0.082
	48	0.032	nd	0.046	0.080	0.069	0.052	0.068	0.058	0.043	0.086	0.059
Lake 56	3	0.148	nd	0.227	0.242	0.316	0.296	0.463	nd	0.304	0.488	0.311
	6	0.107	nd	0.187	0.179	0.226	0.223	0.296	nd	0.225	0.318	0.220
	12	0.077	nd	0.155	0.133	0.162	0.168	0.189	nd	0.167	0.208	0.157
	24	0.055	nd	0.128	0.098	0.116	0.126	0.121	nd	0.124	0.136	0.113
	48	0.040	nd	0.106	0.073	0.083	0.095	0.077	nd	0.092	0.089	0.082
Lake 520	3	0.153	nd	0.393	0.504	0.473	0.453	0.326	0.677	0.246	0.262	0.387
	6	0.111	nd	0.263	0.251	0.254	0.271	0.213	0.355	0.179	0.177	0.230
	12	0.081	nd	0.175	0.124	0.136	0.162	0.139	0.186	0.130	0.119	0.139
	24	0.058	nd	0.117	0.062	0.073	0.097	0.091	0.098	0.094	0.081	0.086
	48	0.043	nd	0.078	0.031	0.039	0.058	0.060	0.051	0.069	0.054	0.054

Table 4.4 Results of a random block design ANOVA testing whether there is a lake effect (random) or time effect (fixed) on TEP-attached bacterial densities for particles in different mean size classes during the summer of 2006.

	ESD (μm)	N	df	F-Stat	p-value
Lake Effect	3	26	2	4.7912	0.0246
	6	26	2	5.6245	0.0150
	12	26	2	7.8738	0.0046
	24	26	2	8.4806	0.0034
	48	26	2	6.6267	0.0087
Time Effect	3	26	8	1.0304	0.4558
	6	26	8	1.6672	0.1874
	12	26	8	2.9505	0.0338
	24	26	8	2.7150	0.0455
	48	26	8	1.4759	0.2455

Table 4.5 **Coefficient of determination for chemical and biological variables plotted against TEP-attached, Free-living, Other-Attached Bacteria, and Total bacterial abundances. A + sign indicates a positive relationship, a – sign indicates a negative relationship. *, **, ***, indicates significance at the alpha = 0.05, 0.01, 0.001 levels respectively.**

Variable	TEP-attached Bacteria				Free-living Bacteria				OA-Bacteria				Total Bacteria			
	520	56	129	All	520	56	129	All	520	56	129	All	520	56	129	All
TEP	+0.55 *	+0.01	+0.15	+0.20 *	+0.09	-0.05	+0.17	0	+0.57 *	+0.17	-0.05	+0.51 ***	+0.61 *	+0.03	0	+0.39 ***
TDOC	-0.31	+0.02	-0.09	0	-0.75 **	+0.01	+0.18	+0.21 *	-0.36	+0.03	0	0	-0.49 *	0	0	+0.03
TDCHO	-0.21	+0.21	+0.15	+0.01	-0.51 *	+0.22	+0.41	+0.07	-0.30	+0.02	-0.70 **	0	-0.39	+0.08	-0.52 *	0
Mono	0	+0.02	+0.25	+0.04	-0.12	+0.46	+0.16	+0.07	-0.01	+0.25	-0.22	+0.05	-0.02	+0.11	-0.08	+0.04
Poly	-0.35	+0.36	0	0	-0.40	-0.09	+0.39	0	-0.45 *	-0.34	-0.50 *	-0.13	-0.55 *	0	-0.64 *	-0.03
CDOC	+0.53 *	0	+0.13	+0.10	+0.31	+0.01	+0.33	+0.06	+0.81 ***	+0.24	0	+0.40 ***	+0.88 ***	0	+0.11	+0.26 **
Chl a	-0.37	0	+0.33	0	-0.31	-0.04	+0.18	-0.04	-0.49 *	+0.09	-0.01	0	-0.56 *	0	+0.05	0
TSS	0	+0.02	+0.15	+0.04	+0.08	-0.11	+0.38	+0.01	+0.29	0	-0.12	+0.28 **	+0.26	0	+0.02	+0.27 **
POC	+0.28	+0.02	0	+0.04	+0.35	-0.03	+0.36	+0.04	+0.60 *	0	+0.02	+0.13	+0.66 *	0	+0.11	+0.13
TEP_{carbon}	+0.81 ***	+0.12	+0.63 **	+0.51 ***	+0.21	-0.01	+0.23	+0.01	+0.56 *	+0.08	-0.12	+0.40 ***	+0.66 **	+0.03	+0.01	+0.36 ***

Table 4.6 **Coefficient of determination for chemical and biological variables plotted against TEP-attached bacterial density (# cells/μm^2) for size intervals as indicated by mean ESD. A + sign indicates a positive relationship, a – sign indicates a negative relationship. *, **, ***, indicates significance at the alpha = 0.05, 0.01, 0.001 levels respectively.**

Variable	Size (μm)	520	56	129	All Lakes
TEP	3	0	-0.21	- 0.10	-0.04
	6	0	-0.35	-0.34	-0.08
	12	+0.06	-0.56*	-0.49*	-0.13
	24	+0.17	-0.71**	-0.44	-0.14
	48	+0.19	-0.65*	-0.37	-0.11
TEP_{carbon}	3	-0.07	-0.33	0	-0.14
	6	-0.03	-0.45	-0.14	-0.18*
	12	0	-0.60*	-0.39	-0.20*
	24	+0.07	-0.65*	-0.47*	-0.15
	48	+0.15	-0.51*	-0.47*	-0.09
Chl a	3	0	-0.04	-0.12	-0.06
	6	+0.02	-0.02	-0.38	-0.14
	12	+0.03	0	- 0.56*	- 0.25**
	24	+0.03	+0.17	- 0.54*	- 0.29**
	48	+0.02	+0.42	- 0.48*	- 0.23*
TSS	3	0	-0.20	-0.20	-0.06
	6	0	-0.25	-0.45	-0.10
	12	0	-0.30	-0.59*	-0.12
	24	0	-0.31	-0.53*	-0.10
	48	0	-0.25	-0.45	-0.07
POC	3	+0.03	-0.48	-0.45	-0.08
	6	0	- 0.63*	- 0.60*	-0.14
	12	-0.01	- 0.83**	- 0.52*	- 0.21*
	24	-0.07	- 0.92***	-0.36	- 0.22*
	48	-0.12	- 0.76**	-0.25	-0.19*
TDOC	3	+0.03	- 0.53*	-0.11	0 (24)
	6	+0.05	- 0.64*	-0.30	+0.01
	12	+0.07	- 0.73**	-0.34	+0.07
	24	+0.06	- 0.66*	-0.21	+0.15
	48	+0.03	-0.42	-0.14	+0.17*
TDCHO	3	+0.07	0	-0.16	+0.01
	6	+0.17	+0.02	-0.43	+0.02
	12	+0.38	+0.07	- 0.54*	+0.04
	24	+ 0.48*	+0.16	-0.44	+0.05

	48	+0.32	+0.25	-0.34	+0.05
Polysaccharide	3	0	+ 0.64*	-0.10	+0.07
	6	0	+ 0.66*	-0.33	+0.12
	12	0	+ 0.60*	- 0.47*	+ 0.15*
	24	+0.02	+0.40	-0.43	+0.10
	48	+0.03	+0.15	-0.36	+0.04
Monosaccharide	3	+0.15	-0.16	-0.09	-0.01
	6	+0.30	-0.11	-0.20	-0.01
	12	+0.54*	-0.03	-0.23	0
	24	+0.56*	0	-0.17	0
	48	+0.32	0.09	-0.12	+0.01
CDOC	3	-0.11	- 0.71**	-0.21	- 0.22*
	6	-0.12	- 0.81**	-0.41	- 0.33**
	12	-0.11	- 0.85***	- 0.45*	- 0.39***
	24	-0.04	- 0.70**	-0.36	- 0.29**
	48	0	-0.38	-0.28	-0.15

4.9 References

Alber, M. and I. Valiela. 1994. Production of microbial organic aggregates from macrophyte-derived dissolved organic material. Limnology and Oceanography **39**:37-50.

Alldredge, A. L., U. Passow, and B. E. Logan. 1993. The abundance and significance of a class of large, transparent organic particles in the ocean. Deep-Sea Research Part I-Oceanographic Research Papers **40**:1131-1140.

Artolozaga, I., B. Ayo, A. Latatu, I. Azua, M. Unanue, and J. Iriberri. 2000. Spatial distribution of protists in the presence of macroaggregates in a marine system. Fems Microbiology Ecology **33**:191-196.

Artolozaga, I., M. Valcarcel, B. Ayo, A. Latatu, and J. Iriberri. 2002. Grazing rates of bacterivorous protists inhabiting diverse marine planktonic microenvironments. Limnology and Oceanography **47**:142-150.

Azam, F. 1998. Microbial control of oceanic carbon flux: The plot thickens. Science **280**:694-696.

Azam, F. and F. Malfatti. 2007. Microbial structuring of marine ecosystems. Nature Reviews Microbiology **5**:782-791.

Berninger, U. G., B. J. Finlay, and P. Kuuppoleinikki. 1991. Protozoan control of bacterial abundances in fresh-water. Limnology and Oceanography **36**:139-147.

Bertilsson, S., L. A. Hansson, W. Graneli, and A. Philibert. 2003. Size-selective predation on pelagic microorganisms in Arctic freshwaters. Journal of Plankton Research **25**:621-631.

Blackburn, N., T. Fenchel, and J. Mitchell. 1998. Microscale nutrient patches in planktonic habitats shown by chemotactic bacteria. Science **282**:2254-2256.

Brachvogel, T., B. Schweitzer, and M. Simon. 2001. Dynamics and bacterial colonization of microaggregates in a large mesotrophic lake. Aquatic Microbial Ecology **26**:23-35.

Caron, D. A., P. G. Davis, L. P. Madin, and J. M. Sieburth. 1986. Enrichment of microbial-populations in macroaggregates (marine snow) from surface waters of the North-Atlantic. Journal of Marine Research **44**:543-565.

Carrias, J. F., J. P. Serre, T. Sime-Ngando, and C. Amblard. 2002. Distribution, size, and bacterial colonization of pico- and nano-detrital organic particles (DOP) in two lakes of different trophic status. Limnology and Oceanography **47**:1202-1209.

Cole, J. J., S. Findlay, and M. L. Pace. 1988. Bacterial production in fresh and saltwater ecosystems - A cross-system overview. Marine Ecology-Progress Series **43**:1-10.

Decho, A. W. 1990. Microbial exopolymer secretions in ocean environments - Their role(s) in food webs and marine processes. Oceanography and Marine Biology **28**:73-153.

Delong, E. F., D. G. Franks, and A. L. Alldredge. 1993. Phylogenetic diversity of aggregate-attached vs free-living marine bacterial assemblages. Limnology and Oceanography **38**:924-934.

Febria, C. 2005. Patterns of hydrogen peroxide among lakes of the Mackenzie Delta and potential affects on bacterial production. Simon Fraser university, Burnaby.

Findlay, S., M. L. Pace, D. Lints, J. J. Cole, N. F. Caraco, and B. Peierls. 1991. Weak-coupling of bacterial and algal production in a heterotrophic ecosystem - The Hudson River Estuary. Limnology and Oceanography **36**:268-278.

Gareis, J. 2007. Underwater irradiance attenuation and photobleaching of chromophoric dissolved organic matter in shallow arctic lakes of the Mackenzie Delta, NWT. Simaon Fraser University, Burnaby.

Grossart, H. P., T. Berman, M. Simon, and K. Pohlmann. 1998. Occurrence and microbial dynamics of macroscopic organic aggregates (lake snow) in Lake Kinneret, Israel, in fall. Aquatic Microbial Ecology **14**:59-67.

Hobbie, J. E., T. L. Corliss, and B. J. Peterson. 1983. Seasonal patterns of bacterial abundance in an Arctic lake. Arctic and Alpine Research **15**:253-259.

Kiorboe, T., H. P. Grossart, H. Ploug, and K. Tang. 2002. Mechanisms and rates of bacterial colonization of sinking aggregates. Applied and Environmental Microbiology **68**:3996-4006.

Kiorboe, T., H. P. Grossart, H. Ploug, K. Tang, and B. Auer. 2004. Particle-associated flagellates: swimming patterns, colonization rates, and grazing on attached bacteria. Aquatic Microbial Ecology **35**:141-152.

Kiorboe, T., H. Ploug, and U. H. Thygesen. 2001. Fluid motion and solute distribution around sinking aggregates. I. Small-scale fluxes and heterogeneity of nutrients in the pelagic environment. Marine Ecology-Progress Series **211**:1-13.

Kiorboe, T., K. Tang, H. P. Grossart, and H. Ploug. 2003. Dynamics of microbial communities on marine snow aggregates: Colonization, growth, detachment, and grazing mortality of attached bacteria. Applied and Environmental Microbiology **69**:3036-3047.

Lemarchand, C., L. Jardillier, J. F. Carrias, M. Richardot, D. Debroas, T. Sime-Ngando, and C. Amblard. 2006. Community composition and activity of prokaryotes associated to detrital particles in two contrasting lake ecosystems. Fems Microbiology Ecology **57**:442-451.

Lesack, L. F. W., P. Marsh, and R. E. Hecky. 1998. Spatial and temporal dynamics of major solute chemistry among Mackenzie Delta lakes. Limnology and Oceanography **43**:1530-1543.

Long, R. A. and F. Azam. 1996. Abundant protein-containing particles in the sea. Aquatic Microbial Ecology **10**:213-221.

Mari, X. and T. Kiorboe. 1996. Abundance, size distribution and bacterial colonization of transparent exopolymeric particles (TEP) during spring in the Kattegat. Journal of Plankton Research **18**:969-986.

Marsh, P. 1986. Modelling water levels for a lake in the Mackenzie Delta. IN Cold Regions Hydrology Symposium, American Water Resources, Association, Fairbanks, Alaska:p.23-29.

Mostajir, B., J. R. Dolan, and F. Rassoulzadegan. 1995. A simple method for the quantification of a class of labile marine pico- and nano-sized detritus: DAPI Yellow Particles (DYP). Aquatic Microbial Ecology **9**:259-266.

Obrien, W. J., A. E. Hershey, J. E. Hobbie, M. A. Hullar, G. W. Kipphut, M. C. Miller, B. Moller, and J. R. Vestal. 1992. Control mechanisms of Arctic lake ecosystems - A limnocorral experiment. Hydrobiologia **240**:143-188.

Passow, U. 2002a. Transparent exopolymer particles (TEP) in aquatic environments. Progress in Oceanography **55**:287-333.

Passow, U. and A. L. Alldredge. 1994. Distribution, size and bacterial-colonization of transparent exopolymer particles (tep) in the ocean. Marine Ecology-Progress Series **113**:185-198.

Riedel, A. 2002. Zooplankton composition and control of heterotrophic flagellates among lakes of the Mackenzie Delta. Simon Fraser University, Burnaby.

Schuster, S. and G. J. Herndl. 1995. Formation and significance of transparent exopolymeric particles in the Northern Adriatic Sea. Marine Ecology-Progress Series **124**:227-236.

Seymour, J. R., J. G. Mitchell, L. Pearson, and R. L. Waters. 2000. Heterogeneity in bacterioplankton abundance from 4.5 millimetre resolution sampling. Aquatic Microbial Ecology **22**:143-153.

Seymour, J. R., J. G. Mitchell, and L. Seuront. 2004. Microscale heterogeneity in the activity of coastal bacterioplankton communities. Aquatic Microbial Ecology **35**:1-16.

Sigee, D. C. 2005. Freshwater Microbiology. Wiley.

Simek, K. and V. Straskrabova. 1992. Bacterioplankton production and protozoan bacterivory in a mesotrophic reservoir. Journal of Plankton Research **14**:773-787.

Simon, M., H. P. Grossart, B. Schweitzer, and H. Ploug. 2002. Microbial ecology of organic aggregates in aquatic ecosystems. Aquatic Microbial Ecology **28**:175-211.

Smith, D. C., M. Simon, A. L. Alldredge, and F. Azam. 1992. Intense hydrolytic enzyme-activity on marine aggregates and implications for rapid particle dissolution. Nature **359**:139-142.

Sommaruga, R. and R. D. Robarts. 1997. The significance of autotrophic and heterotrophic picoplankton in hypertrophic ecosystems. Fems Microbiology Ecology **24**:187-200.

Spears, B. M. and L. F. W. Lesack. 2006. Bacterioplankton production, abundance, and nutrient limitation among lakes of the Mackenzie Delta (western Canadian arctic). Canadian Journal of Fisheries and Aquatic Sciences **63**:845-857.

Squires, M. M. and L. F. W. Lesack. 2003a. Spatial and temporal patterns of light attenuation among lakes of the Mackenzie Delta. Freshwater Biology **48**:1-20.

Tang, K. W., H. P. Grossart, E. M. Yam, G. A. Jackson, H. W. Ducklowl, and T. Kiorboe. 2006. Mesocosm study of particle dynamics and control of particle-associated bacteria by flagellate grazing. Marine Ecology-Progress Series **325**:15-27.

Teichreb, C. 1999. Effects of dissolved organic carbon as a bacterial growth substrate and as an ultraviolet-B radiation sunscreen for aquatic microbial foodwebs in Mackenzie Delta lakes, Northwest Territories. Simon Fraser University, Burnaby.

Thornton, D. C. O. 2004. Formation of transparent exopolymeric particles (TEP) from macroalgal detritus. Marine Ecology-Progress Series **282**:1-12.

Worm, J., K. Gustavson, K. Garde, N. H. Borch, and M. Sondergaard. 2001. Functional similarity of attached and free-living bacteria during freshwater phytoplankton blooms. Aquatic Microbial Ecology **25**:103-111.

Worm, J. and M. Sondergaard. 1998a. Alcian blue-stained particles in a eutrophic lake. Journal of Plankton Research **20**:179-186.

Wu, Q. L. L., Y. W. Chen, K. D. Xu, Z. W. Liu, and M. W. Hahn. 2007. Intra-habitat heterogeneity of microbial food web structure under the regime of eutrophication and sediment resuspension in the large subtropical shallow Lake Taihu, China. Hydrobiologia **581**:241-254.

Yoon, W. B. and R. A. Rosson. 1990. Improved method of enumeration of attached bacteria for study of fluctuation in the abundance of attached and free-living bacteria in response to diel variation in seawater turbidity. Applied and Environmental Microbiology **56**:595-600.

Zimmermann-Timm, H. 2002. Characteristics, dynamics and importance of aggregates in rivers - An invited review. International Review of Hydrobiology **87**:197-240.

APPENDIX A: RAW DATA

Lake 520 TEP data, including results from the microscopic method (ppm), spectrophotometric method (µg xanthan equiv/L), and TEP size distribution constants from the power law equation.

	TEP (ppm)	TEP_{carbon} (µg/ml)	k	Log_{10} k	b	TEP (µg xanthan equiv/L)
04-Jun						42771
12-Jun	137.99	51.99	1237342627	9.09	3.31	18083
17-Jun	25.93	1.15	34935000	7.54	3.42	24275
27-Jun	12.49	0.61	7027400	6.85	2.94	8452
3-Jul	51.36	1.49	1642700	6.22	2.02	9435
11-Jul	9.01	0.37	2265200	6.36	2.57	5477
17-Jul	20.55	0.20	686299	5.84	2.42	6604
24-Jul	12.52	0.40	1694500	6.23	2.51	4429
31-Jul	16.90	0.53	4081100	6.61	2.76	3918
6-Aug	13.40	0.39	1940300	6.29	2.57	3715
13-Aug	8.05	0.28	1844100	6.27	2.69	
21-Aug	4.81	0.17	761349	5.88	2.53	
Mean	**28.46**	**5.24**	**5687795**	**6.41**	**2.64**	

Lake 56 TEP data, including results from the microscopic method (ppm), spectrophotometric method (µg xanthan equiv/L), and TEP size distribution constants from the power law equation.

	TEP (ppm)	TEP_{carbon} (µg/ml)	k	Log_{10} k	b	TEP (µg xanthan equiv/L)
04-Jun						49769
12-Jun	96.81	3.24	7910441	6.90	2.30	27518
17-Jun	36.22	1.05	10063833	7.00	2.86	
27-Jun	16.62	0.72	7455337	6.87	2.83	8649
3-Jul	16.58	0.63	3669068	6.56	2.55	9278
11-Jul	18.55	0.75	8977386	6.95	2.96	6369
17-Jul	6.37	0.31	9561099	6.98	3.44	5189
24-Jul	6.41	0.31	6156804	6.79	3.18	6840
31-Jul	18.02	0.40	397514	5.60	1.99	2830
6-Aug	14.12	0.50	1989877	6.30	2.48	5091
13-Aug	6.23	0.26	3263939	6.51	2.98	
21-Aug	11.97	0.52	9519930	6.98	3.13	
Mean	**22.54**	**0.79**	**6105479**	**6.66**	**2.84**	

Lake 129 TEP data, including results from the microscopic method (ppm), spectrophotometric method (µg xanthan equiv/L), and TEP size distribution constants from the power law equation.

	TEP (ppm)	TEP_{carbon} (µg/ml)	k	Log_{10} k	b	TEP (µg xanthan equiv/L)
04-Jun						44737
12-Jun	295.82	2.57	4864615	6.69	2.06	25710
17-Jun	60.12	1.55	5553555	6.74	2.33	37248
27-Jun	47.58	1.37	10567830	7.02	2.68	9749
3-Jul	18.80	1.37	4830046	6.68	2.80	8059
11-Jul	24.97	0.86	1462869	6.17	2.01	6211
17-Jul	13.01	0.53	5124469	6.71	2.86	9435
24-Jul	12.34	0.49	4830046	6.68	2.80	5451
31-Jul	18.77	0.62	1285051	6.11	2.24	6473
6-Aug	11.04	0.48	2969097	6.47	2.57	7037
13-Aug	25.38	0.97	4432690	6.65	2.44	
21-Aug	9.43	0.47	6393679	6.81	2.97	
Mean	**48.84**	**1.03**	**4744933**	**6.60**	**2.57**	

Lake 520 raw data. CDOC values are absorbance at 330nm.

Sampling Date	TEMP (°C)	DO (mg/L)	% DO	pH	Chl a (µg/L)	TSS (mg/L)	POC (mg/L)	TDOC (mg/L)	TDCHO (mgC/L)	Mono (mgC/L)	Poly (mgC/L)	CDOC
04-Jun	5.3	9.8	78.0	7.72	0.40			7.80	0.47	0.41	0.07	0.427
12-Jun	17.0	8.7	91.8	7.95	2.66				0.87	0.54	0.33	0.420
17-Jun	16.5	7.8	81.2	8.11	1.00	3.71	1.21	8.05	0.53	0.31	0.22	0.428
27-Jun	17.3	9.5	99.0	7.43	0.82	4.00	0.70	8.12	0.74	0.46	0.27	0.406
3-Jul	14.1	7.9	77.8	7.99	0.67	6.00	1.06	8.22	0.87	0.69	0.18	0.424
11-Jul	16.4	10.6	104.1	7.60	0.62			8.29	0.71	0.53	0.19	0.378
17-Jul	20.0	12.6	139.0	8.62	1.92	8.87	1.18	8.41	0.80	0.53	0.27	0.349
24-Jul	16.4	11.3	117.0	8.40	1.82	3.43	0.81	8.17	0.81	0.56	0.25	0.334
31-Jul	20.5	11.8	134.1	8.88	2.35	2.13	0.59	8.61	0.90	0.55	0.35	0.323
6-Aug	20.8	11.4	130.0	8.99	3.34			8.85	0.87	0.51	0.37	0.318
13-Aug	17.0	11.4	118.5	8.89	2.48	2.22	0.68	8.74	0.87	0.51	0.35	0.327
21-Aug	16.3	14.0	142.8	9.06	3.79	1.60	0.47	8.83	0.94	0.53	0.41	0.332
Mean	**16.5**	**10.6**	**109.4**	**8.30**	**1.95**	**4.00**	**0.84**	**8.43**	**0.81**	**0.52**	**0.29**	**0.372**

Lake 56 Seasonal data. CDOC values are absorbance at 330nm.

Sampling Date	TEMP (°C)	DO (mg/L)	% DO	pH	Chl a (µg/L)	TSS (mg/L)	POC (mg/L)	TDOC (mg/L)	TDCHO (mgC/L)	Mono (mgC/L)	Poly (mgC/L)	CDOC
04-Jun	8.9	12.1	102.0	7.50	0.65	79.20	3.20	9.79	1.15	0.83	0.32	0.583
12-Jun	17.8	8.8	93.0	8.06	8.00	30.40	2.26		0.65	0.38	0.27	0.549
17-Jun	15.7	9.1	92.0	8.19	5.69	7.71	2.01	10.34	0.70	0.48	0.23	0.518
27-Jun	17.1	13.2	136.8	8.23	2.63	4.29	1.21	10.11	0.81	0.52	0.29	0.442
3-Jul	13.9	11.2	109.0	8.88	1.92	2.75	0.70	9.91	1.06	0.79	0.27	0.418
11-Jul	17.9	11.9	126.7	10.60	0.89	5.56	1.36	9.82	1.02	0.68	0.34	0.379
17-Jul	19.8	12.7	141.0	8.67	1.22	9.86	1.01	9.66	0.65	0.36	0.29	0.353
24-Jul	15.7	10.6	107.8	10.11	0.84	4.00	0.47	9.49	0.88	0.54	0.34	0.340
31-Jul	20.9	11.0	126.2	10.19	1.08	1.38	0.53	9.52	0.80	0.47	0.34	0.303
6-Aug	20.2	10.7	120.4	10.11	1.40	5.44	0.84	9.57	0.86	0.45	0.41	0.290
13-Aug	16.2	11.7	120.5	9.68	1.08	2.00	0.63	9.77	0.80	0.44	0.35	0.298
21-Aug	14.9	12.5	123.2	10.08	1.23	3.36	0.60	9.68	0.97	0.46	0.51	0.289
Mean	**16.6**	**11.3**	**116.6**	**9.19**	**2.22**	**13.00**	**1.23**	**9.79**	**0.86**	**0.53**	**0.33**	**0.397**

Lake 129 Seasonal data. CDOC values are absorbance at 330nm.

Sampling Date	TEMP (°C)	DO (mg/L)	% DO	pH	Chl a (µg/L)	TSS (mg/L)	POC (mg/L)	TDOC (mg/L)	TDCHO (mgC/L)	Mono (mgC/L)	Poly (mgC/L)	CDOC
04-Jun	5.8	10.6	85.3	7.76	0.43	42.40	2.26	9.00	0.76	0.62	0.14	0.537
12-Jun	14.6	9.6	95.1	7.86	2.16	27.14	2.01	8.93	0.92	0.63	0.29	0.525
17-Jun	15.0	10.5	105.1	8.04	4.32	11.43	1.34		1.03	0.70	0.33	0.527
27-Jun	16.9	11.7	119.0	7.95	5.87	9.43	1.75	8.43	0.77	0.58	0.19	0.444
3-Jul	14.3	8.5	83.5	7.80	3.83	5.43	0.81	8.27	0.86	0.67	0.18	0.439
11-Jul	17.2	9.7	102.1	7.53	1.71			8.42	0.90	0.70	0.20	0.417
17-Jul	19.2	10.2	112.0	8.17	1.80	4.38	0.88	8.66	0.56	0.37	0.19	0.398
24-Jul	16.0	10.7	110.2	8.20	2.58	3.57	0.74	8.53	0.80	0.56	0.24	0.381
31-Jul	20.4	10.5	119.0	8.38	1.86	2.62	0.65	8.65	0.70	0.50	0.20	0.356
6-Aug	20.5	9.3	106.0	8.45	1.68	4.00	0.73	8.59	0.83	0.48	0.35	0.331
13-Aug	16.6	10.0	103.9	8.63	1.56	3.33	0.31	8.33	0.82	0.53	0.29	0.313
21-Aug	15.6	10.5	106.7	8.62	1.61	2.80	0.38	7.55	0.69	0.50	0.19	0.271
Mean	**16.0**	**10.2**	**104.0**	**8.12**	**2.45**	**10.59**	**1.08**	**8.49**	**0.80**	**0.57**	**0.23**	**0.411**

Lake 520 bacteria data. All values except size distribution constants are # cells x 10^5/ml.

	TEP-Attached Bacteria	Free-living Bacteria	Other-Attached Bacteria	Total Bacteria	Size distribution Constants	
					a	b
04-Jun						
12-Jun						
17-Jun	12.02	15.62	91.58	119.22	0.25	0.46
27-Jun						
3-Jul	12.74	8.85	91.97	113.56	0.75	0.58
11-Jul	7.08	7.49	62.13	76.71	1.53	1.01
17-Jul	2.60	7.54	67.19	77.33	1.27	0.90
24-Jul	5.77	15.39	13.38	34.54	1.02	0.74
31-Jul	7.36	4.69	42.24	54.29	0.64	0.61
6-Aug	8.48	4.79	32.21	45.49	1.88	0.93
13-Aug	2.99	3.64	23.61	30.23	0.41	0.46
21-Aug	1.57	4.81	31.78	38.15	0.49	0.57
Mean	**6.73**	**8.09**	**50.68**	**65.50**	**0.91**	**0.70**

Lake 56 bacteria data. All values except size distribution constants are # cells x 10^5/ml.

	TEP-Attached Bacteria	Free-living Bacteria	Other-Attached Bacteria	Total Bacteria	Size distribution Constants	
					a	b
04-Jun						
12-Jun						
17-Jun	7.53	10.23	70.00	87.76	0.25	0.48
27-Jun						
3-Jul	7.36	27.52	88.59	123.47	0.31	0.28
11-Jul	9.52	18.99	53.94	82.45	0.39	0.43
17-Jul	6.62	10.14	59.45	76.21	0.54	0.48
24-Jul	5.81	17.33	64.86	88.01	0.46	0.41
31-Jul	3.65	8.31	76.93	88.89	0.94	0.65
6-Aug				143.34		
13-Aug	4.19	23.89	41.49	69.57	0.49	0.43
21-Aug	14.97	6.38	47.36	68.71	0.96	0.61
Mean	**7.46**	**15.35**	**62.83**	**92.05**	**0.54**	**0.47**

Lake 129 bacteria data. All values except size distribution constants are # cells x 10^5/ml.

	TEP-Attached Bacteria	Free-living Bacteria	Other-Attached Bacteria	Total Bacteria	Size distribution Constants a	b
04-Jun						
12-Jun						
17-Jun	9.89	12.92	144.73	167.55	0.32	0.59
27-Jun						
3-Jul	14.39	11.02	60.03	85.44	0.47	0.60
11-Jul	6.65	10.60	59.33	76.58	0.29	0.33
17-Jul	6.39	8.01	79.04	93.44	0.41	0.46
24-Jul	5.29	6.56	64.88	76.72	0.35	0.49
31-Jul	3.78	3.61	79.98	87.37	0.29	0.38
6-Aug				68.17	0.28	0.41
13-Aug	11.54	7.48	57.40	76.41	0.67	0.71
21-Aug	6.75	2.52	72.12	81.38	0.36	0.37
Mean	**8.08**	**7.84**	**77.19**	**90.34**	**0.38**	**0.48**