Seasonal fluctuations in water quality at four sites on the eastern shore of Lake Winnipeg

Elisa Allegro

Supervised by: Dr. Eva Pip

A thesis submitted in partial fulfillment of the Honours Thesis Course (05.4111/6)

Department of Biology

The University of Winnipeg

2008

In Memoriam

This work is dedicated to the memory of Dr. Ann Smigel, whose generous contribution through the University of Winnipeg Foundation funded a large part of this study. Dr. Smigel will be remembered for her lifetime of service as an isolated teacher in the one-room schools of backcountry rural Manitoba, her keen interest in Lake Winnipeg and environmental and social progress, and her devotion to women's issues and the Ukrainian community, through which she became aware of, and wished to support, Dr. Pip's research

Abstract

Eutrophication of Lake Winnipeg has become a serious concern, but comprehensive water quality data are scarce. The endemic and endangered Lake Winnipeg Physa (*Physella winnipegensis*, Pip, 2004) (LWP) is threatened with extinction as contamination in the lake proceeds. Four sites on the east side of Lake Winnipeg were studied during the ice-free 2007 season. Two of these sites were the only remaining LWP habitats and two were adjacent sites where they can no longer be found. Temperature, pH, total dissolved solids, total suspended solids, total alkalinity, chloride, sulphate, nitrate-N, molybdenum reactive phosphorus (MRP), dissolved organic matter index (DOMI), cadmium, lead, copper, total and fecal coliforms, and cyanobacterial toxins were monitored on a weekly basis. The highest nitrate-N values occurred at the beginning of the season, while MRP was highest during the second half. Stepwise multiple regression yielded a predictive model for total coliforms using DOMI, the nitrate-N: MRP ratio (NPR), and temperature. Correlations were demonstrated between algal toxins and both the preceding MRP and NPR, and concurrent nitrate-N levels. The results of this study underline the need for more rigorous management strategies to address the complex issues that influence the water quality of Lake Winnipeg.

Acknowledgments

I would first like to thank Dr. Eva Pip for her commitment and unwavering support to the completion of this progress. Her dedication to water resources was an inspiration and I am grateful to have experienced working with her. Thanks to my committee members Dr. A.R. Westwood and Dr. J.P.C. Franck for their patience and valuable input. Thank you to Dr. G.E.E. Moodie for his contributions and guidance throughout the duration of the project. The incredible assistance and knowledge from Brenda Vanderkerkhove, Linda Buchanan, and Tara Powell from the University of Winnipeg Biology Department were invaluable throughout the course of the project. Funding provided by Manitoba Hydro and from Dr. Ann Smigel through the University of Winnipeg Foundation made a large part of this research possible. I am exceptionally thankful to Alexander Thomas for his tremendous assistance with the sampling. Finally I would like to thank Nicole Mercier and Liliane Allegro for their constant support and involvement for the completion of this study.

Table of Contents

In Memoriam.....ii

Abstract.....iii

Acknowledgments.....iv

Table of Contents.....v

List of Tables.....vii

List of Appendices.....ix

List of Abbreviations.....xi

1.0 Introduction.....1

2.0 Materials and Method.....5

2.1 Site description.....5

2.2 Field sampling.....5

2.3 Analytical procedures for chemical parameters.....6

2.4 Metal analysis.....7

2.5 Microbiological analysis.....8

2.6 Algal toxins.....8

2.7 Statistical analysis.....10

3.0 Results.....11

3.1 Chemical and Physical Parameter Analysis.....11

3.1.1 Seasonal Fluctuations.....11

3.1.2. Chemical Parameter Intercorrelation Analysis.....15

3.1.3. Principle Components Analysis.....17

3.1.4. NPR Analysis.....17

3.2 Heavy Metals.....18

3.3 Coliform Bacteria.....20

3.4 Cyanobacterial toxins.....23

3.5 LWP Habitat.....23

4.0 Discussion.....23

5.0 Conclusions.....36

6.0 References.....37

List of Tables

Table 1. APHA (1995) methods used for the analysis of filtered water samples from Lake Winnipeg collected during the 2007 ice-free season.....7

Table 2. Calibration regression formulae derived for nitrate-N, MRP, and sulphate analyses.....7

Table 3. Settings for stripping anodic voltametry.....8

Table 4. % Cross-reactivity of polyclonal antibodies with algal toxins.....9

Table 5. Summary of the physical and chemical parameters for Lake Winnipeg water samples pooled for the four sites on the eastern shore during the 2007 ice-free season 11

Table 6. Significant correlations (r and p) among the physical and chemical parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms. Superscripts indicate individual site numbers with significant correlations 16

Table 7. Significant correlations (r and p) between NPR values and selected parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.....18

Table 8. Summary of the heavy metal properties for Lake Winnipeg water samples taken at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....18

Table 9. Significant correlations (r and p) among heavy metals values and selected parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.....20

Table 10. Significant correlations (r and p) between each of total coliform and fecal coliform counts at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.....21

Table 11. Significant correlations (r and p) between the FC/TC ratio and the physical and chemical parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.....22

List of Appendices

Appendix I. Location of site pairs on the east side of Lake Winnipeg sampled during the 2007 ice-free season.....44

Appendix II. Site pairs A (Site 1) and B (Site 2) at Sunset Beach and C (Site 3) and D (Site 4) at Victoria Beach sampled during the 2007 ice-free season.....45

Appendix III. Seasonal fluctuations in temperature at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....46

Appendix IV. Seasonal fluctuation in TSS at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....46

Appendix V. Seasonal fluctuations in TDS at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....47

Appendix VI. Seasonal fluctuations in pH at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....47

Appendix VII. Seasonal fluctuations in total alkalinity at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....48

Appendix VIII. Seasonal fluctuations in chloride at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....48

Appendix IX. Seasonal fluctuations in sulphate at four site on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....49

Appendix X-A. Seasonal fluctuations of nitrate-N, MRP, and NPR at Sites 1 and 2 on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....50

Appendix X-B. Seasonal fluctuations of nitrate-N, MRP, and NPR at Sites 3 and 4 on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....51

Appendix XI. Seasonal fluctuations in DOMI at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....52

Appendix XII. Similarities of chemical parameters in terms of the first three rotated principle components.....52

Appendix XIII. Seasonal fluctuations of cadmium at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....53

Appendix XIV. Seasonal fluctuations of lead at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....53

Appendix XV. Seasonal fluctuations of copper at four sites on the eastern shore of Lake Winnipeg during the ice-free 2007 season.....54

Appendix XVI. Seasonal fluctuations in total coliform counts at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....54

Appendix XVII. Seasonal fluctuations in fecal colifom counts at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....55

Appendix XVIII. Seasonal fluctuations in cyanobacterial toxins at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.....55

List of Abbreviations

Abbreviation	Definition
APHA	American Public Health Association
COSEWIC	Committee on the Status of Endangered Wildlife in Canada
DOMI	Dissolved organic matter index (i.e. absorbance at 275nm)
FC	Fecal coliforms
IUCN	International Union for the Conservation of Nature
LWP	Lake Winnipeg Physa
LWSB	Lake Winnipeg Stewardship Board
MCEC	Manitoba Clean Environment Commission
MPN	Most probable number
MRP	Molybdenum reactive phosphorus
NPR	Nitrate-N:MPR ratio
PCB	Polychlorinated biphenyl
TC	Total coliforms
TDS	Total dissolved solids
TN:TP ratio	Total nitrogen: total phosphorus ratio
TSS	Total suspended solids

1.0 Introduction

Lake Winnipeg is the 10th largest lake in the world and spans an area of 24,389 square kilometers, making it the largest lake in Manitoba (LWSB, 2006). The lake plays a vital role in Manitoba's economy, including tourism, recreation, cottage development, fishing, and hydroelectric generation. The Lake Winnipeg watershed extends for approximately 953 000 square kilometers, covering an area from the Rocky Mountains in Alberta to northwestern Ontario and south into the United States. The watershed supports eight million people and 20 million livestock. Substances produced and released into surface waters within the watershed are eventually deposited into Lake Winnipeg (LWSB, 2006). The watershed includes the Red River which joins the Assiniboine River in Winnipeg. The Red River drains portions of Minnesota, and North and South Dakota, as well as south central Manitoba before water is released into Lake Winnipeg. The Saskatchewan River empties into Lake Winnipeg at Grand Rapids and contributes drainage from Alberta and Saskatchewan (Betcher *et al.*, 1995). The Souris River, which drains into the Assiniboine River, also receives run-off from northwestern North Dakota and southeastern Saskatchewan. The Winnipeg, Saskatchewan, and Red rivers account for 45%, 26%, and 11% respectively of mean monthly flow into Lake Winnipeg (LWSB, 2005). The newly imposed Devils Lake diversion outlet in North Dakota has also raised concerns regarding water quality in the Red River basin. Increased levels of total suspended solids (TDS), sulphate, and salt concentrations, as well as the introduction of foreign organisms from this project have been identified as looming issues with the potential to affect Manitoba surface waters (Manitoba Water Stewardship, 2001).

The decline of surface water quality in the watershed is increasingly being attributed to a number of anthropogenic sources, such as urban effluent, faulty septic

systems, industrial discharges, drainage of wetlands, mining, intensive livestock operations, poor fertilizer practices, land clearing for agriculture, logging, and hydroelectric development (Pip, 2005). To date government attention has focused on nutrient loading and increased nitrogen and phosphorus contamination in the watershed (Jones and Armstrong, 2001). It has been estimated that 29% of the total nitrogen contribution within Manitoba originates from direct municipal and industrial discharges, and 71% has been attributed to watershed processes (particularly agricultural sources). For total phosphorus, 25% originates from municipal and industrial discharges, and 75% stems primarily from agricultural activities (Bourne *et al.*, 2002). Increases in nutrient loading have been thought to alter ecosystem processes in lakes and to exacerbate the growth of bacteria and toxin producing blue-green algae (cyanobacteria) (Chambers *et al.*, 2001).

Increasing frequencies of public beach closures have raised public awareness of bacterial contamination of Lake Winnipeg and have impacted on local economies ranging from tourism to recreational use. Epidemiological studies have linked gastrointestinal and upper respiratory illnesses with recreational water contaminated with pathogenic bacteria (Health and Welfare Canada, 1992). Toxic cyanobacteria secrete neurotoxins, hepatotoxins, lipopolysaccharide toxins, and skin irritants (Health Canada, 2008). Toxic algal blooms have the potential to affect the lake ecosystem as well as the fishing industry, and may pose health threats to those who swim or consume the lake water, or inhale volatile toxins (Health and Welfare Canada, 1992). Toxins may also bioaccumulate in body tissues of aquatic biota such as snails (Lance *et al.,* 2006), freshwater mussels (Watanabe *et al.*, 1997), and fish (Li and Liu, 2003; Codd *et al.*, 1997 *in* Health Canada, 2008).

In Manitoba urban effluent, agriculture, forest clear cutting practices, and pulp mill effluent have been associated with increased levels of TDS, nitrate-N, total suspended solids (TSS), dissolved organic matter index (DOMI), and phosphorus in surface waters (Pip, 2006; Pip, 2005). Winnipeg's water treatment also includes the addition orthophosphate to tap water to control lead leached from the distribution system, which eventually contributes to phosphorus loading in Lake Winnipeg. Increased TDS, nitrate-N and metal levels have been attributed to the mining industry as well, since there are many such operations on the Precambrian Shield (Pip, 2005). Chemicals such as organochlorine insecticides and polychlorinated biphenyls (PCB) are also being found in the south basin of Lake Winnipeg (Rawn *et al.*, 2000). Flooding events in the Red River Valley further exacerbate the contamination problem as they accelerate the transport of materials originating from landfills, sewage lagoons, livestock operations, and hazardous wastes as well as chemical and fuel storage sites (Stewart *et al.*, 2000).

In the face of these concerns is a lack of seasonal information on water quality for Lake Winnipeg, especially data specific for near-shore littoral communities. The littoral zone provides a refuge for open water organisms at different stages of their life histories, supports a wide variety of plants and invertebrates (Moss, 1998), and provides important spawning habitat for fish. This area has the greatest amount of biodiversity and is the most productive area of a lake. However it is also the zone in which contaminants first enter the lake ecosystem, and therefore the impact of pollutants is the most intense in this interface.

Within the littoral zone, gastropods are a vital component of the aquatic food web and its energy flow. They feed on periphyton and macrophytes, and in turn form the food for higher trophic levels. All of the 26 gastropod species found in Lake Winnipeg occur

in this zone (Pip, 2006). Freshwater gastropods are disproportionally represented as 20% of all organisms on IUCN's Red List of Threatened Species, and yet only comprise 5% of all gastropod fauna (Strong *et al.*, 2008). The Lake Winnipeg Physa, *Physella winnipegensis* Pip, 2004 (LWP), endemic to Lake Winnipeg, has been classified endangered since 2002 (COSEWIC, 2002). Genetic studies using DNA sequencing have found that the LWP is phylogenetically distinct from other Physa snails in central Canada (Pip and Franck, 2008). The LWP is found on submerged algae-covered rocks exposed to wave action at depths of < 1m (Pip, 2004). The LWP habitat is found in the near-shore littoral zone where contaminated inputs and human disturbances are the most acute.

Freshwater mussels have also declined from 11 species to 5 in Lake Winnipeg, and at present 98% of the freshwater mussel communities in the lake are composed of only the two most tolerant species (Pip, 2000; Pip, 2006). Freshwater mussels are the most endangered group of animals in North America. These organisms have long life spans (up to 100 years) and therefore can accumulate pollutants to extreme levels (Pip, 1995).

The primary objective of this study was to investigate whether water quality predictors can be utilized to model algal toxins and coliform bacteria, which are important for public health as well as indicators of water quality. A secondary objective was to obtain detailed information about habitat quality at the last two remaining LWP sites and compare them with two adjacent sites where the LWP have not been found since 2001 (Pip, unpublished).

2.0 Materials and Methods

2.1 Site description

Four sites on the east shore of the southern basin of Lake Winnipeg (Appendix I) were examined during the ice-free 6 May – 21 October, 2007 season. These sites were grouped as two pairs: one pair (Sites 1 and 2) in the Sunset Beach area (50° 31'N, 96° 36'W) and the other pair (Sites 3 and 4) at Victoria Beach (50° 42'N, 96° 34'W). Sites 1 and 3 consisted of the last two remaining LWP habitats while Sites 2 and 4 (Appendix I) were adjacent locations where this species no longer occurs (Pip, unpublished data). All four sites were exposed sand beaches interspaced with boulders, which normally form the microhabitat for LWP. All four sites were also subject to extensive cottage development and recreational use.

Site 1 (Appendix II-A) featured beach-front cottages, some of which had pipes carrying drainage water directly onto the beach. Site 2 (Appendix II-B) received drainage from a marshy area which in turn received wastewater effluent from a sewage lagoon. Site 1 was separated from Site 2 by a groin which also served as a boat launch. Site 3 (Appendix II-C) was located southeast of the Victoria Beach marina. The intake for the water treatment plant for the community of Victoria Beach was located adjacent to Site 4 (Appendix II-D).

2.2 Field sampling

The four sites were sampled for water at weekly intervals from 6 May, 2007 (immediately after ice-off) to 26 August, 2007. Two more supplementary sampling sessions occurred at four-week intervals on 23 September and 21 October, giving a total of 19 visits to each site. The lake froze over soon afterwards.

All water samples were collected 10cm below the surface and placed on ice. Water temperature was read directly using a thermometer *in situ*, as was pH using a portable pH meter 29 (Radiometer, Copenhagen) calibrated on site. One L of lake water was filtered on site through a pre-weighed Whatman No. 1 filter paper which was immediately refrigerated and subsequently frozen at -20°C. These filters were then air-dried and weighed to quantify TSS. The filtrate collected from this procedure was frozen within 12 hours at -20°C as three samples and tested for: (1) nitrate-N, DOMI, and molybdenum reactive phosphorus (MRP), (2), sulphate, chloride, total alkalinity, and TDS, and (3) dissolved cadmium, lead and copper. For sample (3), metal-free high-density polyethylene bottles were used for sample collection (VWR TraceClean, West Chester, Pennsylvania).

A whole (un-filtered) sample (4) was collected in a sterile bottle (VWR International, Mississauga, Ontario) and immediately refrigerated, then returned to the laboratory for total coliform (TC) and fecal coliform (FC) quantification. Each week, samples (4) were collected at the same time of day for each respective site. Another whole water sample (5) was collected, immediately placed on ice, and subsequently frozen at -20°C. for algal toxin analysis.

2.3 Analytical procedures for chemical parameters

Chemical parameters in samples (1) and (2) were analyzed using standard analytical methods recommended by the American Public Health Association (1995) (Table 1). TDS was measured using a TDSTesr 1 (Oakton, Wards Natural Science, St. Catharine's, Ontario). Spectrophotometry was conducted using a Pharmacia LKB Ultrospec III (Pharmacia LKD Biochrom, Science Park, Cambridge, England). DOMI was estimated using ultraviolet screening of acidified samples at 275 nm (e.g. Pip, 2005).

Table 1: APHA (1995) methods used for the analysis of filtered water samples from Lake Winnipeg collected during the 2007 ice-free season.

Parameter	Analytical Procedure
TDS	Potentiometery
NO_3-N	UV Spectrophotometry
DOMI	UV Spectrophotometery
MRP	Spectrophotometry
Sulphate	Turbidimetry
Chloride	Colorimetric Titration
Total Alkalinity	Potentiometric Titration

Regression formulae for calibration curves for nitrate-N, MRP and sulphate are given in Table 2.

Table 2: Calibration regression formulae derived for nitrate-N, MRP, and sulphate analyses.

Parameter	Formula
Nitrate-N	mg/L NO_3-N= 3.62 (corrected absorbance at 220nm)*+0.01 $r = 0.997$, $p<0.0001$
MRP	mg/L PO_4-P = 1.69 (absorbance at 880 nm)+ 0.006 $r = 0.9995$, $p < 0.00001$
Sulphate	mg/L SO_4 = 99.61 (absorbance at 420 nm)+ 7.36 $r = 0.93$, $p<0.001$

* Corrected absorbance= (absorbance at 220 nm) – 2(absorbance at 275 nm)

2.4 Metal analysis

Cadmium, lead, and copper were analyzed in sample (3). Stripping anodic voltametry was used. This method allows for direct analysis of raw water samples, eliminating solvent extraction steps, and thus minimizing losses (Mann and Lintern, 1984). Settings for the non-overlapping voltage windows during the plating and stripping process (Pip, 2000) are shown in Table 3.

Table 3: Settings for stripping anodic voltametry.

Stage	Voltage (mV)	Time (sec)
Pre-circulate	-	5
Plating	-1000	60
Hold	-900	15
Sweep 375 cycles/sec	Cd-900- -550	1
	Pb-550- -375	1
	Cu-375- -50	1
Strip	0	2
Rest	-100	Finish

For this method, three 5 mL replicate aliquots of thawed filtrate were each plated four times for the three metals. The standard additions method (Mann *et al.*, 1974) was applied to compensate for matrix absorption effects, using atomic absorption metal standards (Fisher Scientific Co., Fair Lawn, New Jersey). Three sequential additions were used for each aliquot after plating the un-spiked samples. Therefore each sample was plated a total of 12 times.

2.5 Microbiological analysis

Sample (4) was used to inoculate media within 24 hr. of collection. Employing the Durham tube method, lactose and Brilliant Green media were inoculated in five-fold serial dilutions and incubated at 37°C. and 44.5°C respectively for 24 h. TC and FC were recorded as Most Probable Number (MPN) estimates (Cappuccino and Sherman, 2005).

2.6 Algal toxins

Sample (5) was used to quantify algal toxins. The sample was subjected to three rapid freeze-thaw cycles, then filtered through a GF/C glass microfiber filter (1.2 μm) (Lehman, 2007). Duplicates of the filtrate were analyzed by ELISA (Lehman, 2007)

using polyclonal antibodies to bind with microcystins (Beacon Analytical Systems Inc., Portland, Maine). This method does not require solvent extraction and is sensitive to low microcystin concentrations (Pyo *et al.*, 2005), but distinguishes different toxin variants to different degrees (Table 4).

Table 4: % Cross-reactivity of polyclonal antibodies with algal toxins.

Algal Toxin Variant	% cross-reactivity
Microcyctin-LR	100
Microcystin-YR	87
Microcystin-RR	48
Nodularin	31

After incubation with the antibody solution, excess microcystin antigens linked to peroxidase were bound to any remaining un-bound antibody sites (Lehman, 2007). The bound peroxidase activity was analyzed spectrophotometrically as a color reaction at 450 and 605 nm using a Beckman DU-7 spectrophotometer (Beckman Instruments, Irvine California). The relationship between toxin concentration and absorbance was reciprocal (Lehmann, 2007). The absorbance at 605 nm was subtracted from that at 450 nm to give the corrected absorbance (CA), which was multiplied by 100.

The %Bo was calculated as:

$$\%Bo = (CA) \times 100 / \text{Average absorbance of negative controls}$$

The negative controls consisted of antibody tubes that contained no algal toxin. The %Bo was calculated for all unknown samples, calibration standards and quality control

samples. The resulting regression formula for the calibration standards (over a range of 0 – 5.0 µg/L microcystin-LR) was (R = -0.91, p <<0.0001, n = 35):

Log toxin concentration (µg/L) = -0.0225 (%Bo) + 1.435

2.7 Statistical analysis

The data were pre-tested (Sokal and Rohlf, 1981) to determine whether distributions were appropriate for each statistical test. Significant ANOVA tests were post-tested using Tukey's Honestly Significantly Different tests (HSD) to identify specifically where the significant differences existed. All tests, including multivariate analyses (Tatsuoka 1971, Williamson 1972) were performed using SPSS (Chicago, Illinois). Deletion criteria for multiple regression analysis were, p in =0.050 p out =0.100. The critical significance level for all tests was strictly $p = 0.050$. Since $p = 0.051$ is less significant $p = 0.049$, in this study p values ≤ 0.050 were considered "marginal", while $p = 0.051$ was rejected as not significant (Gregory, 2004), because of the increased likelihood of α – error due to chance of values in the peri-significant zone (Dickson and Massey, 1957; Pip, 1978)

3.0 Results

3.1 Chemical and Physical Parameter Analysis

3.1.1 Seasonal Fluctuations

The means, standard errors and ranges for the chemical and physical parameters for all sites sampled in the 2007 season can be found in Table 5.

Table 5: Summary of the physical and chemical parameters for Lake Winnipeg water samples pooled for the four sites on the eastern shore during the 2007 ice-free season.

Parameter	Mean ± S.E.	Range
TSS (mg/L)	74 ± 12	11 - 705
TDS (mg/L)	228.16 ± 9	60 - 390
pH	7.5 ± <0.01	6.8 - 7.9
Total alkalinity. ($mgCaCO_3/L$)	140 ± 7	50 - 430
Chloride (mg/L)	14.7 ± 0.9	1 - 41
Sulphate (mg/L)	33 ± 2	7 - 70
Nitrate-N (mg/L)	0.37 ± 0.04	<0.01 - 2.22
MRP (mg/L)	0.08 ± 0.01	<0.01 - 0.34
DOMI (275 nm)	0.215 ± 0.006	0.112 - 0.362
Temperature (°C)	18 ± 0.6	7 - 27

Appendix III shows temperature patterns during the ice-free season at the four sites. The distribution was expected with cooler temperatures occurring during the early spring and fall. The highest temperatures occurred from the end of June until August, with the exception of a slight drop occurring 1 July. ANOVA showed no significant differences in temperature distributions among the four sites.

TSS measurements (Appendix IV) peaked at the beginning of the season on 20 May at Sites 1, 2, and 3, but then stayed between 11 and 130 mg/L until 22 July where the value at Site 1 peaked to a maximum of 705 mg/L. TSS rose again at Sites 3, 2, and 1 on 5 Aug. Site 1 had the highest overall values, while Site 4 had the lowest. Tukey-HSD

tests showed that concentrations of TSS at Site 1 were significantly different than those at Site 4, which had the lowest mean for TSS. Mean TSS decreased sequentially from Sites 1 through 4 reflecting the degrees of turbulence and wave exposure at these sites. Seasonal TSS fluctuations for Sites 2 and 3 were significantly correlated (Pearson r = 0.78, p <0.001, n = 16).

Sites 1 and 2 had higher TDS values from May until the beginning of June and mid-July until the end of August (Appendix V). A peak was observed at all sites on 24 June. The highest readings of the season occurred on 22 July, where Sites 1 and 2 reached 390 and 360 mg/L respectively. During the last sampling session all site values were between 140 and 190 mg/L. Tukey-HSD tests showed that Sites 1 and 2 had significantly higher concentrations than Sites 3 and 4. TDS values were correlated at Sites 1 and 2 (r = 0.96, p < 0.001, n =19), and at Sites 3 and 4 (r = 0.90, p <0.001, n = 19). Sites 2 and 3 were also correlated (r = 0.52 and p = 0.012, n = 19).

Values for pH in the month of May started between 7.4 and 7.6, except at Site 2 which commenced at 7.9 (Appendix VI). Site 2 had the lowest values among the four sites. All sites fell below 7.0 in September and October. Tukey-HSD tests indicated no significant differences among the four sites. Seasonal patterns were not significantly correlated among any of the sites.

Total alkalinity was initially high at Sites 4, 1, and 3, but then dropped after the first sampling week (Appendix VII). The seasonal fluctuations at Sites 1 and 3 (r = 0.52, p = 0.12, n=19), 1 and 4 (r = 0.40, p = 0.044, n=19), and 3 and 4 (r = 0.46, p = 0.025, n=19) were significantly correlated. Site 2 on the other hand increased in alkalinity from 130 to 430 mgCaO_3/L during the first three weeks and had the highest value of total alkalinity of all the four sites. Tukey-HSD multiple range tests showed that overall values

at Site 2 were significantly higher than at Sites 3 and 4.

Chloride concentrations had a similar pattern to that of total alkalinity, where values at Sites 1, 3, and 4 decreased after the first sampling session (Appendix VIII). Among these three sites, Sites 1 and 3 ($r = 0.59, p = 0.004, n=19$), 1 and 4 ($r = 0.49, p = 0.017, n=19$), and 3 and 4 ($r = 0.65, p = 0.001, n=19$) were significantly correlated. The chloride concentration at Site 2 increased on 20 May and had the highest value of 41 mg/L. Tukey-HSD multiple range tests showed that values at Sites 1 and 2 were significantly higher than at Sites 3 and 4.

Sulphate levels increased during mid-May, especially at Site 1 which reached a maximum of 70mg/L (Appendix IX). Levels fluctuated erratically for much of the season. Tukey-HSD tests showed no significant differences in sulphate ranges among the four sites. The fluctuations at Sites 1 and 2 ($r = 0.57, p = 0.005, n=19$), 2 and 3 ($r = 0.42, p = 0.036, n=19$), and 3 and 4 ($r = 0.66, p = 0.001, n=19$) were significantly correlated.

Nitrate-N concentrations were highest at the beginning of May and then fell after the first sampling session (Appendix X: A-B). Another peak is evident on 1 July at all sites which occurred after a heavy rainfall on 26 June. Nitrate-N levels then showed a general decrease until the end of the season. Tukey-HSD tests indicated no significant differences in ranges of concentration among the four sites. The fluctuations in concentrations at Sites 1 and 2 ($r = 0.89, p = <0.001, n=19$), 2 and 3 ($r = 0.55, p = 0.008, n=19$), 2 and 4 ($r = 0.49, p = 0.016, n=19$), and 3 and 4 ($r = 0.94, p < 0.001, n=19$) were significantly correlated.

MRP levels on the other hand were relatively low and then steadily climbed (Appendix X: A-B) until freeze-up. There were also peaks in MRP concentration around 1 July, coinciding with those of nitrate-N. MRP concentrations peaked again on 5 August

and remained high for the rest of the season, especially at Site 4 on 23 September where a maximum concentration of 0.34 mg/L was reached. Therefore phosphorous loadings in these sites were the highest during the later portion of the season. As with nitrate-N, ANOVA and Tukey-HSD tests revealed no significant differences in ranges of concentration among the four sites. Sites 1 and 2 ($r = 0.89$, $p < 0.001$, n=19), 1 and 3 ($r = 0.63$, $p = 0.002$, n=19), and 2 and 3 ($r = 0.65$, $p = 0.001$, n=19) were significantly correlated throughout the season.

The associated nitrogen/phosphorus ratios (NPR) can also bee seen in Appendix X (A-B). NPR started with higher values at the beginning of the season, peaking at different times, but then gradually decreased. The peaks in NPR are more pronounced at Sites 3 and 4, especially around 22 July. NPR ranged from 0.07-33.3, but ANOVA revealed no significant differences among the four sites overall. The end of season NPRs dropped at all sites, especially at Sites 3 and 4 where the values decreased below 1.

DOMI values were generally higher at Sites 1 and 2 than Sites 3 and 4, although a very sharp peak in DOMI occurred on 17 June at Site 3 (Appendix XI). Tukey-HSD showed no significant differences in ranges of concentration among the four sites. Correlation analysis revealed that only Sites 1 and 2 ($r = 0.46$, $p = 0.023$, n=19) were significantly seasonally correlated

3.1.2. Chemical Parameter Intercorrelation Analysis

Analysis of the chemical data from all sites revealed a number of highly significant correlations (Table 6). Most of these correlations were non-linear and yielded improved regression fits when one or both parameters were transformed as natural logarithms. Total alkalinity, chloride and sulphate each showed temporal correlations with 7 other chemical parameters. Highly significant relationships were apparent among TDS, sulphate, chloride, and total alkalinity. These correlations were also significant at all four individual sites when the data were analyzed separately, except for the correlations between TDS and chloride which were only significant at Sites 2, 3, and 4. The only inverse correlations were between MRP and pH (Table 6), and between nitrate-N and ln temperature (r = -0.39, p <0.001, n=76).

Table 6: Significant correlations (r and p) among the physical and chemical parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms. Superscripts indicate individual site numbers with significant correlations.

	TSS	TDS	pH	Total Alk (TA)	Chloride (Cl)	Sulphate (Su)	NO_3-N	MRP	DOMI
TSS	X	$0.56^{(1,2,4)}$ <0.001 lnTSS	NS	$0.44^{(2,4)}$ <0.001 lnTA, lnTSS	$0.51^{(2,3)}$ <0.001 lnTSS	$0.37^{(1)}$ <0.001 lnTSS	$NS^{(3,4)}$	$0.29^{(3)}$ 0.010 lnTSS	$NS^{(1)}$
TDS		X	NS	$0.74^{(1,2,3,4)}$ <0.001 lnTA, lnTDS	$0.79^{(2,3,4)}$ <0.001 lnCl, lnTDS	$0.83^{(1,2,3,4)}$ <0.001 lnTDS, lnSu	$0.40^{(1,3,4)}$ <0.001 lnNO_3 -N	0.21 0.033 lnPO_4-P	$0.23^{(3)}$ 0.025
pH			X	NS	$NS^{(2)}$	NS	$0.20^{(1,2,4)}$ 0.045 lnNO_3 -N	$-0.41^{(-3,-4)}$ <0.001 lnPO_4-P	NS
Total Alk				X	$0.70^{(1,2,3,4)}$ <0.001 lnTA	$0.63^{(1,2,3,4)}$ <0.001 lnTA, lnSu	$0.42^{(1,3,4)}$ <0.001	$0.28^{(1)}$ 0.008 lnPO_4-P	$0.38^{(2)}$ <0.001
Chloride (Cl)					X	$0.59^{(2,3,4)}$ <0.001 lnCl ,lnSu	$0.38^{(2,3)}$ <0.001 lnNO_3-N	$0.24^{(4)}$ 0.019 lnCl, lnPO_4-P	0.23 0.024
Sulphate (Su)						X	$0.21^{(1,4)}$ 0.032 lnNO_3-N lnSu	NS	NS
NO_3-N							X	$0.23^{(2)}$ 0.023	$NS^{(3)}$
MRP								X	$0.43^{(2)}$ <0.001
DOMI									X

3.1.3. Principle Components Analysis

Seasonal fluctuation patterns were further investigated with principle components analysis to compare the seasonal similarities among the 9 water chemistry parameters listed in Table 6. KMO (Kaiser-Meyer-Olkin) measure of sampling adequacy and Bartlett's tests of sphericity indicated that factor analysis was suitable, since correlations between pairs of variables could not be explained by the other variables (Kaiser, 1974). Convergence was reached after six iterations; Kaiser normalization in the rotation phase was applied. This resulted in the extraction of 3 primary eigenvalues which accounted for 38.9%, 17.9%, and 12.9% respectively in the entire data block, thus representing a total cumulative percentage of 69.7%. Scree plots (Cattell, 1966) of the eigenvalues indicated that the remaining principle components each contributed less than 5% of the total variability. The three axes represented by these eigenvalues were rotated orthogonally using the Varimax method (Tatsuoka, 1971). This was done to minimize the number of parameters with high loadings for any individual factor (Pip, 1988a). Appendix XII shows the 3-dimentional interrelationships of fluctuations of the chemical parameters distributed along the three primary eigenvalues. Thus TDS, total alkalinity, chloride, and sulphate clustered together, reflecting their close relationships in Table 6.

3.1.4. NPR Analysis

Table 7 shows the chemical and physical parameters which were correlated with NPR (nitrate-N: MRP ration) values. NPR was highly positively significant with pH, highly inversely correlated with TSS, DOMI, and somewhat less with temperature, and marginally positively with chloride. However nitrate-N values were not correlated with TSS, or DOMI (Table 6).

Table 7: Significant correlations (r and p) between NPR values and selected parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.

	TSS	pH	Chloride	DOMI	Temperature
NPR	-.031, 0.006 lnNPR	0.44, <0.001 lnNPR	0.19, 0.049	-0.28, 0.007	-0.23, 0.021 lnTemp

Stepwise multiple regression analysis using the significantly correlated parameters and ln (NPR) as the dependent variable resulted in the equation below, where $F = 19.2$ and $p < 0.0001$:

$$\ln(\text{NPR}) = 3.07\,(\text{pH}) - 0.10\,(\text{Temperature}) - 19.6$$

The associated β coefficients for pH and temperature were 0.67 and -0.49 respectively, indicating that pH was relatively more important.

3.2 Heavy Metals

The means and ranges for the three metals analyzed can be found in Table 8. Of the three metals, Pb had the largest range from 0.1- 36.3 µg/L.

Table 8: Summary of the heavy metal properties for Lake Winnipeg water samples taken at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.

Metal	Mean ± S.E. (µg/L)	Range
Cd	4.08 ± 0.45	0.1-23.5
Pb	11.82 ± 0.62	0.1-36.3
Cu	9.5 ± 0.41	0.1-19.4

The highest concentrations of Cd occurred at the beginning of May (Appendix XIII). There were fairly large peaks at Sites 4 and 3 on 27 May and 3 June respectively. Site 4 then declined for the rest of the season, while Site 3 experienced additional peaks.

Lead concentrations also varied greatly among the four sites (Appendix XIV). The highest concentrations occurred in May until the beginning of June, and there was another peak in September. Sites 2 and 3 had higher concentrations at the beginning of the season and then dropped, especially at Site 2 on 24 June. Site 2 peaked on July and Site 3 had the maximum value of 36.3 µg/L on 23 September, but then fell by 21 October. Sites 1 and 4 on the other hand started the season with relatively low values and then fluctuated erratically for the remainder. The end of season values for Sites 1 and 4 were relatively high compared to concentrations experienced earlier in the season.

For Cu, values peaked at the end of June and remained high until the beginning of August (Appendix XV). Values at Sites 3 and 4 were generally lower than Sites 1 and 2. Site 1 experienced a maximum value of 19.4 µg/L on June 24 which was also the highest value of the season.

Among the metals, only Pb was significantly correlated with Cu ($r = 0.031$, $p = 0.003$, n=76). When other physical and chemical parameters were included, Pb was significantly correlated with temperature and pH (Table 9). Cu levels were significantly correlated with TDS, sulphate, and chloride.

Table 9: Significant correlations (r and p) among heavy metals values and selected parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.

Metal	TDS	pH	Chloride (Cl)	Sulphate (Su)	Temperature
Cd	NS	NS	NS	NS	NS
Pb	NS	0.25,0.015 lnPb	NS	NS	0.32, 0.002 lnPb
Cu	0.22, 0.029 lnCu, lnTDS	NS	0.20, 0.045	0.21, 0.036 lnCu, lnSu	NS

3.3 Coliform Bacteria

The seasonal fluctuations in TC (Appendix XVI) among the four sites varied considerably. The highest counts occurred at the beginning of July until the end of August. Sites 1 and 2 for the most part had the highest overall values, although Site 3 had a large peak on 29 July. The mean for all sites was 196.5 MPN/100mL (± 35.0 S.E.) and ranged from 1-1600 MPN/100 mL.

FC varied at all sites throughout the season and the highest FC values occurred at Site 2 (Appendix XVII). The highest values were observed at the beginning of the season from May until mid-July. The mean for all four sites was 17.0 MPN/mL (± 3.0 S.E.) and ranged from 0- 170 MPN/100mL.

TC was positively significantly correlated with DOMI, MRP, TSS, total alkalinity and TDS, and negatively with NPR (Table 10). TC also showed an inverse correlation with Pb (r = -0.27, p = 0.008, and n = 76) when TC was ln transformed. FC was also positively significantly correlated with total alkalinity, TDS and MPR, and negatively with pH and NPR. Both TC and FC were more strongly correlated with NPR than with MRP (Table 10).

Table 10: Significant correlations (r and p) between each of total coliform and fecal coliform counts at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.

	TSS	TDS	pH	Total Alk (TA)	Sulphate (Su)	MRP	DOMI	NPR
TC	0.36, 0.002 lnTC lnTSS	0.21, 0.033 lnTC	NS	0.26, 0.010 lnTA	0.21, 0.041 lnTC	0.37, 0.001 lnTC	0.46, <0.001 lnTC	-0.40, <0.001 lnTC
FC	NS	0.21, 0.033	-0.36, 0.001 lnFC	0.24, 0.020 lnTA	NS	0.20, 0.043 lnFC	NS	-0.23, 0.024 lnFC

Stepwise multiple regression using the significantly correlated parameters in Table 10 yielded a significant model with as the ln (TC) as the dependent variable ($F = 10.4$, $p < 0.0001$):

$$\ln TC = 10.45\,(DOMI) + 0.07\,(Temperature) - 0.06\,(NPR) + 1.19$$

The β coefficients for DOMI, NPR and temperature were 0.34, -0.25 and 0.23 respectively, thus DOM had the greatest relative importance. MRP in place of NPR resulted in a relatively less significant model ($F = 10.03$, $p < 0.001$) due to the stronger correlation of TC with NPR ($r = -0.40$, $p < 0.001$) than with MRP ($r = 0.37$, $p = 0.001$). When samples collected in water <10°C were excluded from the data pool, the model further improved ($r = 0.61$, $F = 11.0$, $p < 0.0001$) to:

$$\ln TC = 11.3\,(DOMI) + 0.12\,(Temperature) - 0.07\,(NPR) + 0.10$$

The β coefficients were 0.37, 0.30, and -0.24 for DOMI, temperature, and NPR respectively.

An alternate version of the regression equation was obtained when Pb was included in the list of independent variables. This analysis yielded the following equation ($F = 9.24, p < 0.0001, n = 76$):

$$\ln TC = 9.18\,(DOMI) + 0.07\,(temperature) - 0.06\,(NPR) - 0.06\,(Pb) + 2.08$$

The β coefficients were 0.30, 0.25, -0.25 and -0.21 respectively. Thus the relative contributions of temperature and NPR became equal when Pb was factored into the equation.

LnTC and FC were positively significantly correlated ($r = 0.32, p = 0.002, n = 76$). The FC/TC ratio also showed significant inverse correlations with temperature, TSS, DOMI, and MRP (Table 11).

Table 11: Significant correlations (r and p) between the FC/TC ratio and the physical and chemical parameters at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season (n=76). Where ln values occur, non-linear correlations have been transformed to natural logarithms.

	Temperature	**TSS**	**DOM**	**MRP**	**N/P ratio**
ln FC/TC	- 0.37, 0.001	- 0.32,0.004 ln TSS	- 0.30,0.004	- 0.20,0.039 lnMRP	-0.19, 0.050 lnN/P

3.4 Cyanobacterial toxins

Cyanobacterial toxin levels ranged from 0.01- 1.61 µg/L with a mean of 0.43 µg/L, and were already elevated at the beginning of May. They peaked again during the second week of July but otherwise fluctuations were poorly synchronized at the four sites (Appendix XVIII). Site 3 experienced a maximum recorded concentration of 1.61 µg/L on July 22.

Analysis using cross function correlation demonstrated that cyanobacterial toxins were significantly correlated with the previous MRP levels ($r = 0.22$, $p = 0.029$, $n = 74$) when the last two outlaying sampling dates were excluded. Toxins were also inversely correlated with preceding NPR ($r = -0.21$, $p = 0.033$, NPR ln transformed). Toxin concentrations were correlated with concurrent nitrate-N levels as well ($r = 0.20$, $p = 0.045$, and $n = 76$), although the correlation was less significant than with MRP.

3.5 LWP Habitat

Differences between LWP sites (1 and 3) and non-LWP sites (2 and 4) were compared using paired t-tests. Total alkalinity, DOMI, cadmium, and FC were significantly lower at LWP sites ($p < 0.050$). However TSS values were higher at Sites 1 and 3, reflecting the greater wave exposure and turbulence at these sites.

4.0 Discussion

Lake water chemistry is dependent on many different factors and conditions, as there are complex processes operating which affect the interactions and dynamics of the various parameters. Determining the initial sources of these contaminants in the watershed can be an immense task, but it is essential to understand the behavior of these substances in the environment and their eventual release into Lake Winnipeg when

addressing the lake's declining ecosystem health and water quality.

The parameter values found in the present study were within water chemistry ranges previously reported for central Canada (Pip, 1988b). Previous studies in Lake Winnipeg have also found similar concentrations as well as correlations to those found in this study, such as those between TDS and each of nitrate-N and DOMI (Pip, 2006). The very strong and consistent correlations among TDS, chloride, total alkalinity, and sulphate reflect the common geochemical sources of these substances and their mutually similar behaviors in the near-shore environment. Research conducted on other prairie lakes have also demonstrated these types of associations; when TDS values increased, sulphate and chloride (as well as magnesium) levels increased proportionally (Hammer, 1970). TSS also showed multiple correlations with other parameters in this study suggesting the origin of many of these substances was related at least in part to runoff and turbulence. Many different types of substances are also transported with suspended particles in lake ecosystems. These associations have been employed in water quality modeling (i.e. sediment-transport modules), where chemicals, metals, and also microorganisms circulate with suspended sediments in lake systems (Rajar *et al.,* 1997).

. Internal circulation patterns, particularly seiches and long-shore currents, may influence the eventual deposition of substances transported from the watershed to the near-shore littoral communities. Horizontal gradients of dissolved salt, suspended sediment, and algae have been demonstrated in Lake Winnipeg (Brunskill *et al.*, 1980). Although the circulation patterns in the southern basin are not well understood, it has been thought that water entering via the Red River splits and travels along the west and east shores, before turning inwards on meeting the Winnipeg River (IJC, 2000). A previous study conducted in Manitoba has demonstrated that water quality of surface

waters is also dependent on adjacent land use and human activity (Pip, 2005). Research conducted in the Great Lakes has found that the origins of contaminants within the water bodies were associated with local industries operating within the area (Marvin *et al.*, 2004).

The areas studied were all subject to cottage development and recreational use. Currently more than 10 000 cottages exist around Lake Winnipeg (Pip, 2006). According to the latter author, pressure from growing cottage populations is increasingly impacting on shoreline water quality. More intense cottage development associated with decreasing lot sizes, road construction, and increased drainage are augmenting the amount of runoff of sediments into the near-shore water. As native vegetation is removed and replaced with lawns, which have a much reduced capacity to retain sediments and nutrients, combined with increased fertilizer and pesticide use, greater amounts of contaminants are reaching the lake. Increases in phosphorus are often associated with cottage development, especially in lakes with low alkalinity (Garrison and Wakeman, 2000). Protruding pipes were noted emptying drainage water directly from back yards into the lake at one of the sites studied. Besides contributing nitrogen, phosphorus, and various household chemicals, residential wastewater may also have been treated with water softeners and this can lead to excessive concentrations of sulphate and sodium (Jones *et al.*, 1998). The addition of chlorine as a disinfectant by municipal water treatment plants also has the potential to contaminate near-shore areas of the lake with chlorinated organic compounds.

There are many large-scale industries located in Lake Winnipeg's watershed which contribute to the degradation of water quality, including logging operations on the East side of the lake. Research on the effects of clear-cutting in the vicinity of Precambrian Shield lakes have shown that dissolved organic carbon, chlorophyll,

nitrogen, potassium, chloride, and silicon all increased by 10-40% within two years of experimental clear-cut logging (Steedman, 2000). It was also found that the response of water quality parameters in a lake with little shoreline disturbances and moderate deforestation was comparable to that which occurred in lakes with extensive upland disturbances and shoreline deforestation. In Manitoba, logging operations contribute DOMI, TSS, and phosphorus to surface waters (Pip, 2005). Pulp and paper mills, such as those found on the Winnipeg and Saskatchewan Rivers also contribute organic matter to Lake Winnipeg (COSEWIC, 2002) as well as a variety of other chemicals involved with, pulping, fungal control, de-inking, and bleaching of the products (Thompson *et al.*, 2001). Apart from the dissolved wood materials, the effluent also contains heavy metals, as well as nitrogen and phosphorus. Mining operations in Manitoba have also been found to influence heavy metal levels in surface waters as well as contribute TDS (Pip, 2005). In the same study, Pb was found to be positively correlated with nitrate-N, and Cu was correlated with TDS in the watershed. The average value for Pb in the present study corresponded to levels recorded in 2001 survey on the east side of Lake Winnipeg, but was higher for Cd (Pip, 2006). The latter discrepancy was due to five occurrences Cd concentrations of >10 μg/L recorded at Sites 3 and 4 in 2007. Normally higher metal concentrations in waters on the Precambrian Shield are due to the acidic drainage and increased metal solubilities (Bannatyne and Teller, 1984 in Pip, 2006). Thus, Cd, Pb, and Cu levels are found to be higher on the East side of Lake Winnipeg when compared to the West (Pip, 2006). However, there have been other studies which have found higher metal levels in areas with high pH due to high concentrations of base cations, associated with smelting operations (Blais *et al.*, 1999).

All three metals monitored had substantial peaks at the beginning of the season when spring runoff occurred in the surrounding watershed. Shafer *et al.* (1997) found that filterable and particulate metals increased by up to 5 -17 fold during events like spring melt. Rainfall events have also been associated with increased levels of Cd, Cu, Pb, and Zn, as metals from the surrounding environment would leach into the lake catchment area (Morfett *et al.*, 1988).

When compared to the same 2001 survey conducted on the east side of Lake Winnipeg (Pip, 2005), the 2007 Cu concentrations were not as elevated. The correlation between Cu and sulphate may be indicative, at least in part, of the application of copper sulphate as an algicide, to counteract the problematic algal blooms in cottage areas. The use of this algicide was visibly apparent as shoreline encrustations during the course of this study. The use of this algicide is banned except on private lands where the receiving water body is entirely within the landowner's properly and has no outflow, or where provincial license is granted (e.g. domestic water source). Otherwise illegal private use has been difficult to monitor and enforce. Other sources of Cu in cottage areas may include preservatives leached from docks, decks, and exterior wood construction materials. The City of Winnipeg also recommends the use of copper sulphate to kill tree roots in residential sewer connections. According to sewage services contractors, such domestic use is very common (G. Haliday, personal communication). Copper is highly toxic to aquatic ecosystems and is readily bioaccumulated by freshwater organisms (Pip, 1990a; Pip 1990b). The Canadian Water Quality Guideline for the Protection of Aquatic Life (Health Canada, 1995) is 2-4 μg/L for Cu. Many of the values from the 2007 sampling exceeded this range.

The boat launch and marina at Sites 2 and 3 respectively could also be contributing heavy metals into the surrounding water from substances originating from fuel leaks and spills, marine paints, and fishing equipment. Previous studies using freshwater mussels as biomonitors found that organisms in a sailing boat harbor had accumulated strikingly higher concentrations of Cu, Zn, Pb, and Cd when compared to organisms collected in open water (Balogh, 1988). Agriculture has also been linked to metal contamination. Hog feeds are often enriched with metals, e.g. copper and zinc, which are used as growth promoters and to increase food conversion rates (Wong, 1985),as well as to control disease (MCEC, 2007). Other trace elements that may be present in feed include cadmium, lead, nickel, cobalt, boron, selenium, and iodine (Strauch, 1987). Only 5-10% of the copper and zinc in feed is absorbed by the hogs (Wong, 1985).The excretion of the metals by the animals and subsequent application onto fields, pose the risk of eventually leaching and contaminating ground and surface waters (MCEC, 2007).

Intensified agricultural operations occurring in Lake Winnipeg's watershed, particularly in the southern portion, are increasingly being associated with eutrophication of the lake. The changing land management practices in agriculture have effectively allowed anthropogenic sources of annual nitrogen and phosphorus loading to terrestrial ecosystems to equal all natural sources of these nutrients combined, due to the use of synthetic fertilizers on cropland (MCEC, 2007). Livestock have also been identified as a critical contributor involved in the eutrophication of Manitoba surface waters. Although some regulations have been established for large scale operations, smaller operations have often ignored recommended land management practices and may apply manure, particularly from hogs, to fields regularly throughout the year, which is eventually lost

through snowmelt (MCEC, 2007). It has been estimated that agriculture alone is responsible for 5% of total nitrogen and 15% of total phosphorus loading to Lake Winnipeg (LWSB, 2006).

The highest values of nitrate-N occurred at the beginning of the season. Large amounts of nitrogen occur in spring runoff from agricultural lands. A study by Armstrong (2002) conducted on the Assiniboine River also revealed peaks of total nitrogen loadings during this time.

The relationship between nitrogen and phosphorus in lake ecosystems has been much explored and research has found that at times when low levels of nitrate are present in lake water, the subsequent addition of this nutrient increased the release of phosphorus from sediments (Jensen and Anderson, 1992). In the present study, the influx of nitrate-N on 1 July occurred after a period of relatively low nitrate levels; higher MRP levels followed and continued during the second half of the season. High phosphorus solubilities have been found to be maintained by wind resuspension and bioturbation combined with high pH, as well as anoxic conditions from warm and calm weather, and macrophyte senescence in later parts of the season (Welch and Cooke, 1995).

Nitrate-N and MRP also shared several correlations with other parameters, namely TDS, total alkalinity, and chloride. Biochemical processes involving the assimilation of nitrogen and phosphorus by lake organisms correspond with increases in total alkalinity. Wolf-Gladrow *et al.* (2007) determined that 1 mole of nitrogen, in the form of nitrate or nitrite, leads to a corresponding increase in total alkalinity by 1 mole. N_2 fixation also increases total alkalinity by one mole per mole of N when NH_4^+ is released. The same principle was found for phosphate (H_3PO_4, $H_2PO_4^-$, HPO_4^{2-}, or PO_4^{3-}). According to the latter workers, the increase in total alkalinity from nitrogen utilization was found to be

more substantial as organisms are able to uptake it more readily than inorganic phosphate.

The inverse correlation demonstrated between MRP and pH may be related to the ability of phytoplankton in the water column to produce different alkaline phosphatases, which can be activated at varying pH levels (Olsson, 1990). Organisms produce these enzymes to release orthophosphate from the total phosphorus pool in refractory molecules (Berman, 1970) and contribute to inorganic phosphorus concentrations. Chrost and Overbeck (2005) found that the highest alkaline phosphatase activity occurred during phosphorus depletion and that this activity was not dependent on biomass. Nitrate levels have also been found to influence phosphatase activity in *Anabaena* (cyanobacterium) (Bone, 1971).

The NPR is also important in eutrophic lakes and as this ratio decreases, nitrogen becomes limiting as it does not accumulate in lakes and is readily denitrified by organisms (Moss, 1998). High phosphorus levels and TN: TP ratios have previously been associated with nitrogen limitation (Downing and MacCauley, 1992). A low NPR in a lake ecosystem indicates a high degree of pollution and as eutrophication progresses, the NPR moves toward that found in sewage (Pick and Lean, 1987).

The Guidelines for Recreational Water Quality (Health and Welfare Canada,1992) state that TC counts should not exceed 200 MPN /100mL, but the 2007 results show that this level was exceeded for most of the summer months, especially at Sites 1 and 2. Coliform bacteria belong to the family *Enterobacteriaceae* and constitute 10% of microorganisms present in the intestinal tract of humans and other animals (Prescott *et al.*, 2005). Agriculture has been implicated as being one of the major sources of bacterial contamination within North American watersheds. A study conducted on the Sumas River watershed in British Columbia and Washington State revealed that increasing the number

of livestock animal units over a given area increased fecal coliform counts in freshwater tributaries (Berka *et al.*, 2001).

Although the Guidelines for Canadian Recreational Water Quality (Health and Welfare Canada, 1992) do not state a specific limit for FC counts in recreational water, it is generally accepted that they should not be present in areas where these types of activities are occurring. When contaminated waters are used for recreation, there most certainly exists the possibility for the transmissions of disease (Prescott *et al.*,.2005). FC counts were somewhat high at the four study sites, especially at Site 2. FC have been found to be concentrated in the top 20cm of wet sand cores in beach areas (Alm *et al.*, 2003, Whitman and Nevers, 2003). This can influence the potential for pathogen transmission as people swimming in the lake will always have a period of contact with this zone, especially small children who splash and wade in shallow waters. Beach sand can act as a reservoir for pathogens that have the potential to be recycled into the water column after disturbances. Storms have been found to be responsible for bringing fresh bacteria and suspended materials with runoff into the near-shore areas of a lake from the surrounding catchment (Ackerman and Weisberg, 2003). This supports the rain-based beach water quality warning systems used in some regions. In Lake Winnipeg, Hamblin (1976) found that seiches and storm surges were more extreme in the southern basin than in the larger north basin, although it has not been determined if there are any differences in bacterial contamination between the north and south basins.

Williamson *et al.* (2004) have also found associations with *E. coli* and near-shore water levels, and determined that the density of this bacterium was a function of the departure from mean water levels for the previous five days. These workers also identified birds, namely gulls and terns, among the animal sources of *E. coli*. Bonilla *et*

al. (2007) noted that bird, animal, and human sources, as well as protozoan predation could account for irregular distributions of bacterial populations at Florida marine beaches. A survey conducted at a Chicago beach has also found that when *E. coli* counts were lagged by one day after bird counts, correlations were observed (Whitman and Nevers, 2003). However when analyzed on a daily basis, the presence of birds on the beach was not correlated with *E. coli* concentrations in the sand or water. There were also no significant differences in *E. coli* concentrations between areas with and without gull flocks.

The relationship revealed between TC and the other parameters used in the linear regression models points to the biological and hydrodynamic processes which are occurring and promoting the proliferation of bacterial populations at these study sites. Dissolved organic matter is readily assimilated by bacterial cells as nutrients (Prescott *et al.*, 2005), and increased temperatures stimulate the metabolic activity of these microorganisms (Cappuccino and Sherman, 2005). Also MRP rather than nitrate-N was the more significant factor in relation to TC and FC, as both were positively correlated with MRP and inversely correlated with NPR. In the case of coliform bacteria the correlations with NPR reflected more general contamination conditions. Furthermore heavy metals in the environment are often toxic to bacterial populations, for example Cu (Prescott *et al.*, 2005). In this study, Pb was inversely correlated with TC.

The significant correlations between cyanobacterial toxins and MRP, nitrate-N, and NPR revealed in this study support other research. Nitrate has been correlated with the growth of *Microcystis* and microcystin production (Jiang *et al.*, 2008), and the presence of toxin producing species was highly correlated with total nitrogen (Giani *et al.*, 2005). Yan *et al.* (2004) also found that the production of microcystin-LR was

increased in the presence of nitrate, when compared to other forms of nitrogen such as urea and ammonium. They suggested that this form of inorganic nitrogen is more significantly associated with microcystin-LR production than total nitrogen.

This study showed that MRP levels were more significantly correlated with the toxins than were nitrate levels. Increased total and dissolved phosphorus has been linked to elevated microcystin levels (Wang *et al.*, 2002). In Alberta lakes microcystins were correlated with total phosphorus, and negatively correlated with inorganic nitrogen (Kotak *et al.*, 2000). Although these two nutrients seem to be important with regard to cyanobacterial toxins in water, no consistent relationship has emerged from the research; thus other aspects may be influencing toxin levels in different areas.

The data also revealed a significant inverse correlation between NPR and toxin levels. Pick and Lean (1987) were unable to link low NPR with cyanobacteria, however they did find that TN: TP ratios above 30 decreased the presence of cyanobacteria. Different cyanobacterial genera capable of producing toxins have been found within distinct ranges of TN: TP ratios (Lehman, 2007). Therefore using this aspect of water quality alone to predict toxin levels has been difficult as it may depend on species composition. The inverse correlation between NPR and toxin levels demonstrates the ability of cyanobacteria to fix nitrogen under limiting conditions, as they are the major types organisms involved in the accumulation of nutrients in the absence of nitrogen (Prescott *et al.*, 2005). Orr and Jones (1998) have found that the production of microcystin has been strongly associated with periods of active cell division. Accordingly the increases in algal toxin concentrations result from increased cyanobacterial growth closely succeeding increases in MRP.

Although warmer temperatures are known to influence the growth of cyanobacteria, the presence of algal toxins in at the four sites was not significantly correlated with temperatures throughout the season, which supports the findings of Kotak *et al.* (2000). In fact, the first peak in toxin levels at all sites occurred during the first sampling session at the beginning of the ice-free season. This observation is similar to the findings of Kotak *et al.* (1995) in Alberta lakes. Jones *et al.* (1998 *in* Health Canada, 2008) have also found that microcystin-LR could stay in water long after algal populations have disappeared. Jiang *et al.* (2008) demonstrated that different toxin producing cyanobacteria may be present in the water column at different temperatures.

To remain consistent, samples were taken during the same part of the day, as some blue-green algae can migrate vertically in the water column using buoyancy regulation at different periods of the day (Visser *et al.*, 1997). Kotak *et al.* (1995) found that microcystin production in blooms was elevated during the day possibly because of the effect of light on toxin production. The intensity of light and the production of toxins have been positively correlated up to the point of maximum growth rate, but production is inhibited past this point as light increases (Wiedner *et al.*, 2003).

The use of copper sulphate as an algicide in the littoral zones of Lake Winnipeg may be distorting possible associations between other water quality parameters and algal toxin levels. Other factors such as iron (Jiang *et al.*, 2008), dissolved oxygen, and pH (Prescott *et al.,* 2005; Billam *et al.,* 2006) have also been found to encourage cyanobacterial blooms.

Lake Winnipeg has shown differences in toxin levels at the same sites during different seasons. Gurney and Jones (1997, *in* Health Canada, 2008) reported a maximum of 300 µg/L at Site 4 in 1996, while the maximum at this site during the 2007 season was

0.98 μg/L . This extreme concentration in 1996 was thought to have occurred due to a large algal bloom that was pushed to the near-shore areas by wind. Understanding and monitoring algal toxin concentrations is especially important at this site as it is directly adjacent to the intake of the water treatment plant which supplies the community of Victoria Beach with drinking water.

The deterioration of water quality in littoral areas of Lake Winnipeg is effectively restricting the distribution of the endemic and endangered LWP (COSEWIC, 2002). The many different processes occurring in the watershed and in the local vicinity of their habitat are contributing to their decline. One notable factor at these sites is the continuing alteration of shoreline. The construction of groins in these areas for a boat launch (Site 2) and as breakwaters in recreational areas (Site 4) impedes the turbulence requirement characteristic of the LWP habitat (Pip, 2004). The decreased mixing capacity and interference with longshore currents at these altered sites result in increased concentrations of pollutants (Pip, 2006). During the 2007 season, only four LWP individuals were found at Sites 1 and 3 combined (Pip, unpublished data). This requirement for high energy environments is reflected in the increased levels of TSS at the remaining LWP sites. Gastropods in Lake Winnipeg have also previously been found to be highly inversely correlated with Cd, Pb, and Cu (Pip, 2006), and the LWP were highly sensitive to Pb (Pip, 2004). Understanding the processes that affect the endangered LWP is critical as their distribution has catastrophically declined in Lake Winnipeg in only the last few years. Loss of biodiversity in North American freshwater systems is rapidly declining, and unless the current deleterious practices are ended, the extinction of freshwater species will continue.

5.0 Conclusions

The present study identified phosphorus, nitrogen, dissolved organic matter, as well as TSS as major factors responsible for deterioration water quality in Lake Winnipeg. A number of the water chemistry parameters were significantly correlated, and most of these relationships were non-linear. Copper, lead and cadmium often exceeded guidelines for protection of aquatic life, and may be contributing to loss of biodiversity in near shore areas, including the Lake Winnipeg Physa. This study demonstrated highly significant relationships between total coliform bacteria and dissolved organic matter, temperature, and NPR. Microcystins were correlated with preceding inorganic phosphorus and NPR, and with concurrent nitrate-N levels. Rising anthropogenic pressures in the watershed, including agriculture cottage development, urban effluent, clearcuting, mining, and hydroelectric development are contributing nutrients and various contaminants which ultimately affect water quality of the lake. These sectors must all be addressed in a comprehensive management strategy with the aim of reducing adverse impacts, and hopefully providing an improved outlook for the recovery irreplaceable and economically vital resource.

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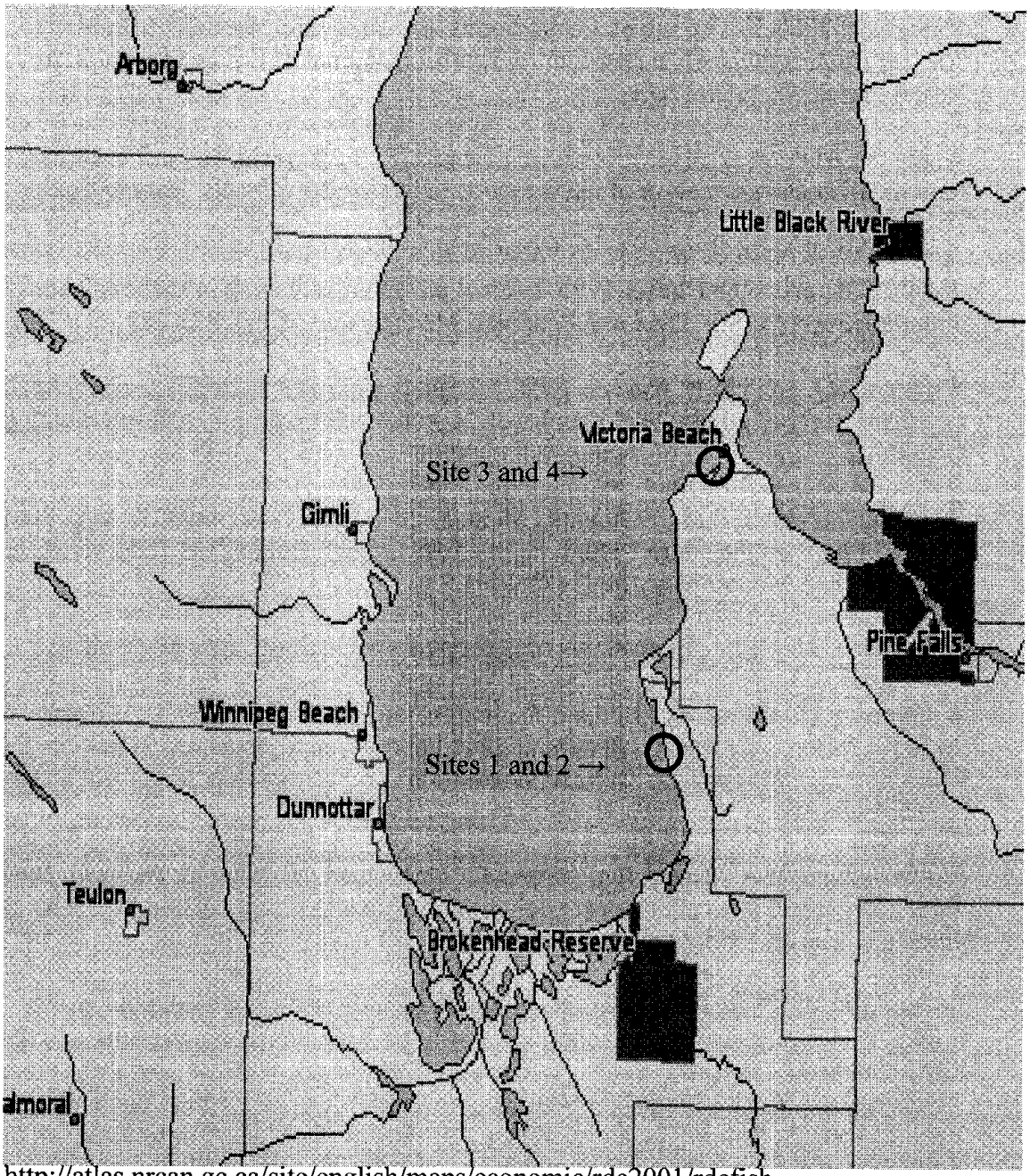
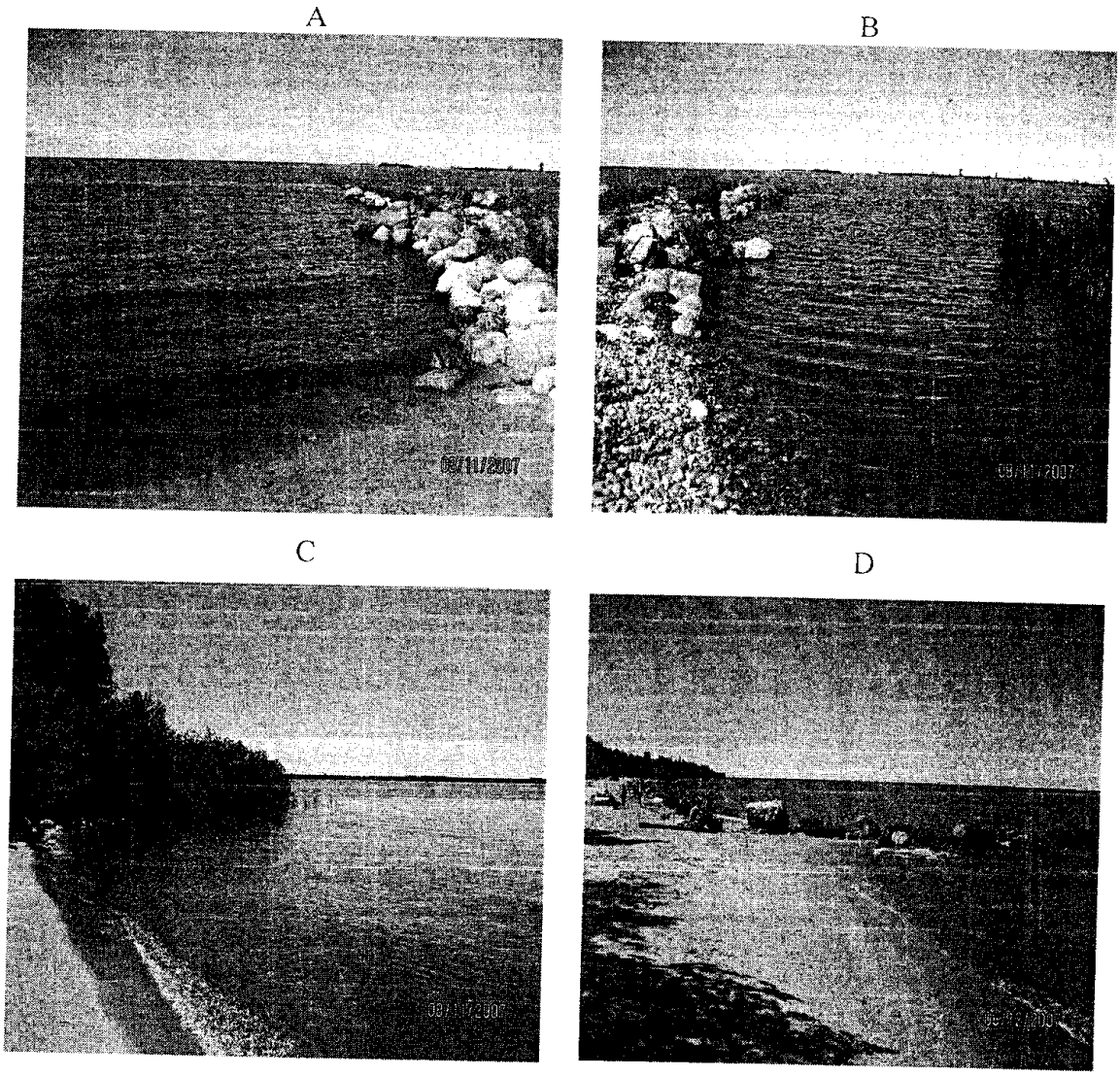


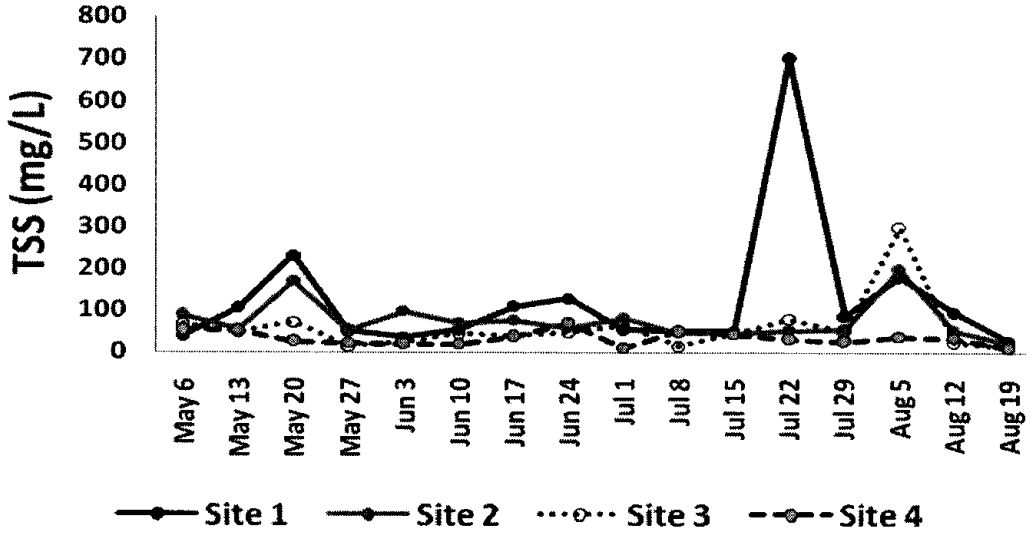
http://atlas.nrcan.gc.ca/site/english/maps/economic/rdc2001/rdcfish

Appendix I: Location of site pairs on the east side of Lake Winnipeg sampled during the 2007 ice-free season.

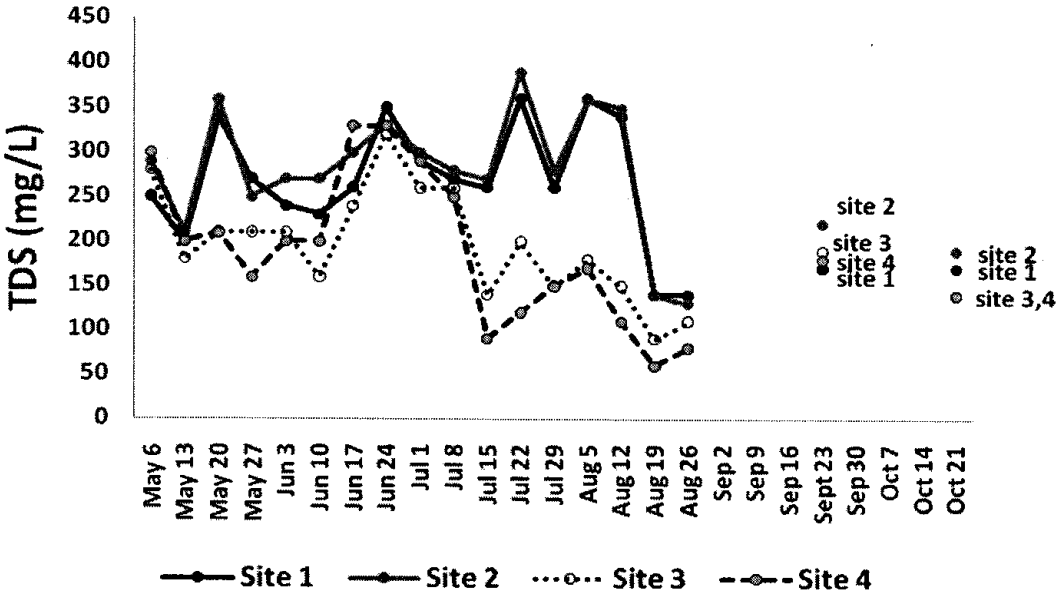


Appendix II: Site pairs A (Site 1) and B (Site 2) at Sunset Beach and C (Site 3) and D (Site 4) at Victoria Beach sampled during the 2007 ice-free season.

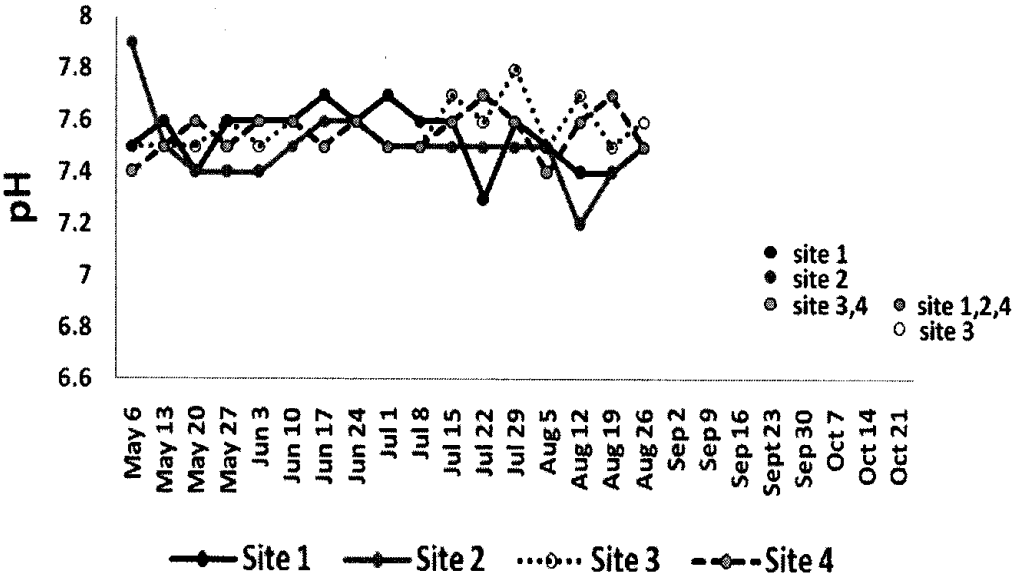
Appendix III: Seasonal fluctuations in temperature at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



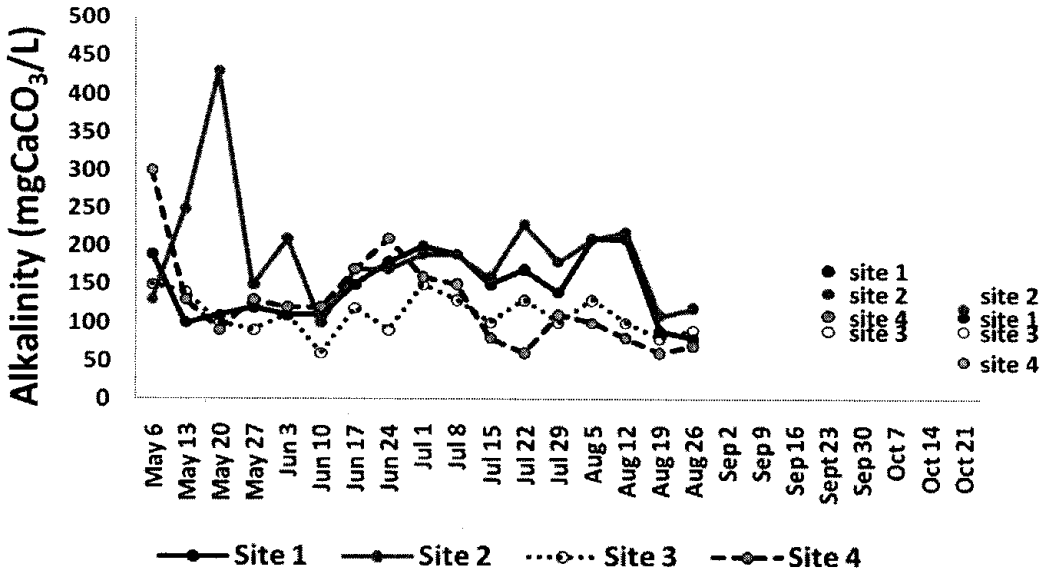
Appendix IV: Seasonal fluctuation in TSS at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



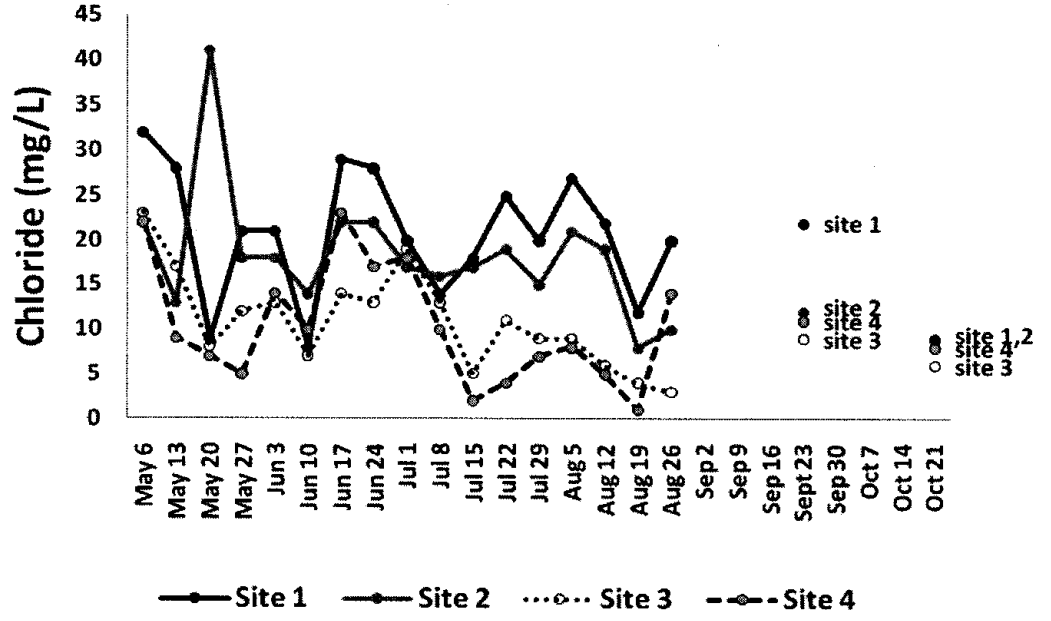
Appendix V: Seasonal fluctuations in TDS at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



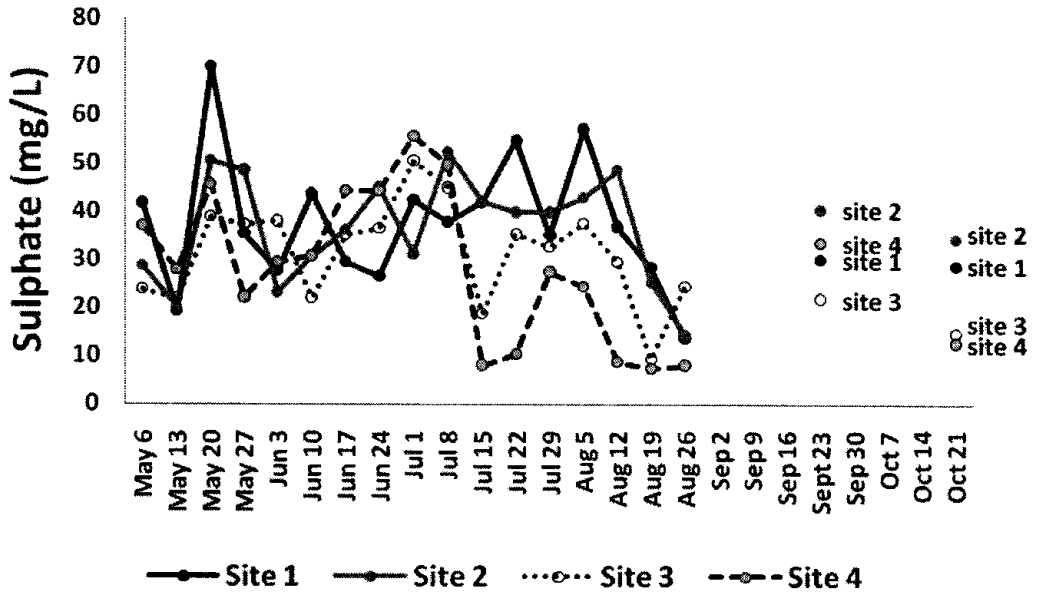
Appendix VI: Seasonal fluctuations in pH at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



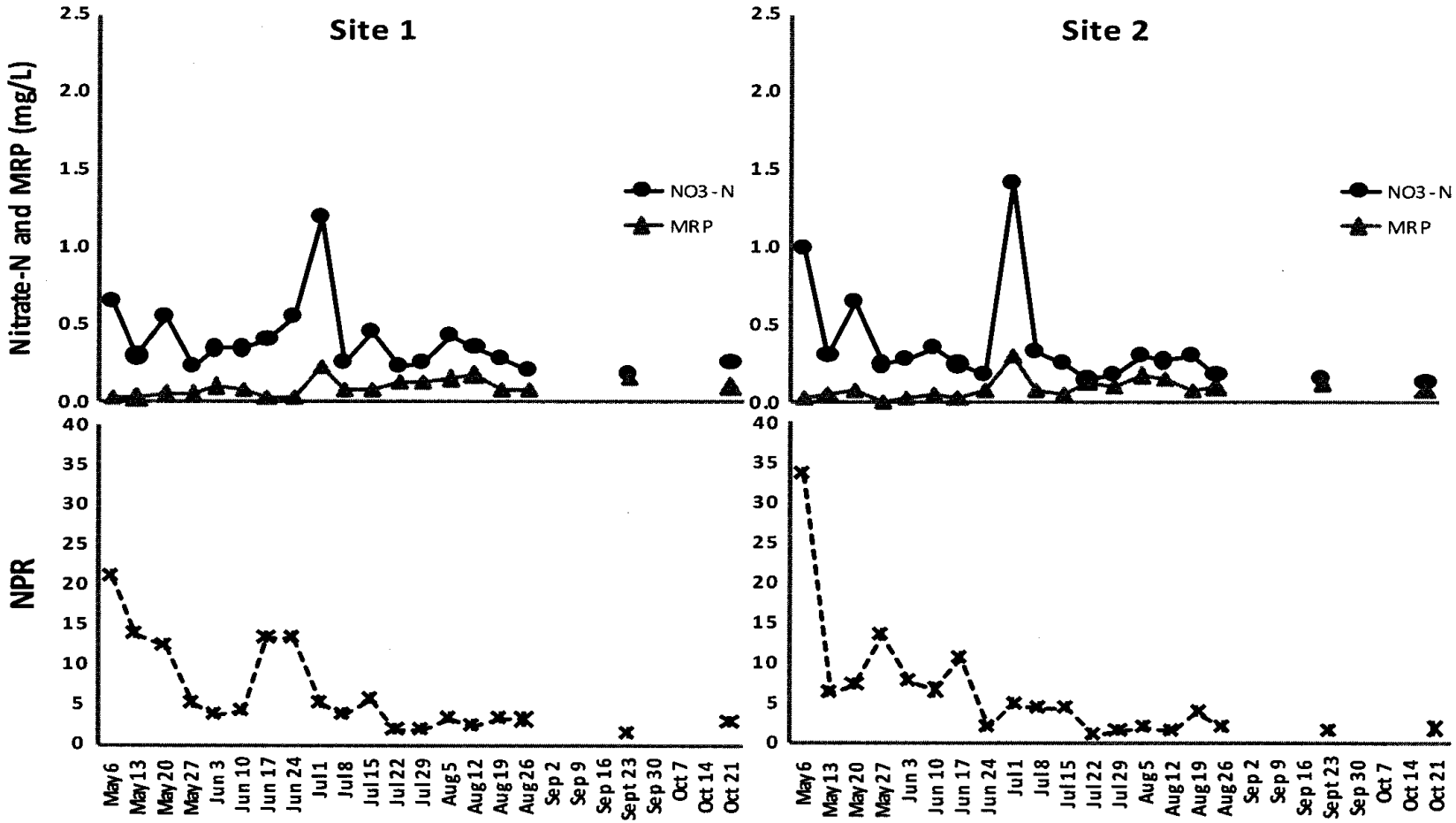
Appendix VII: Seasonal fluctuations in total alkalinity at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



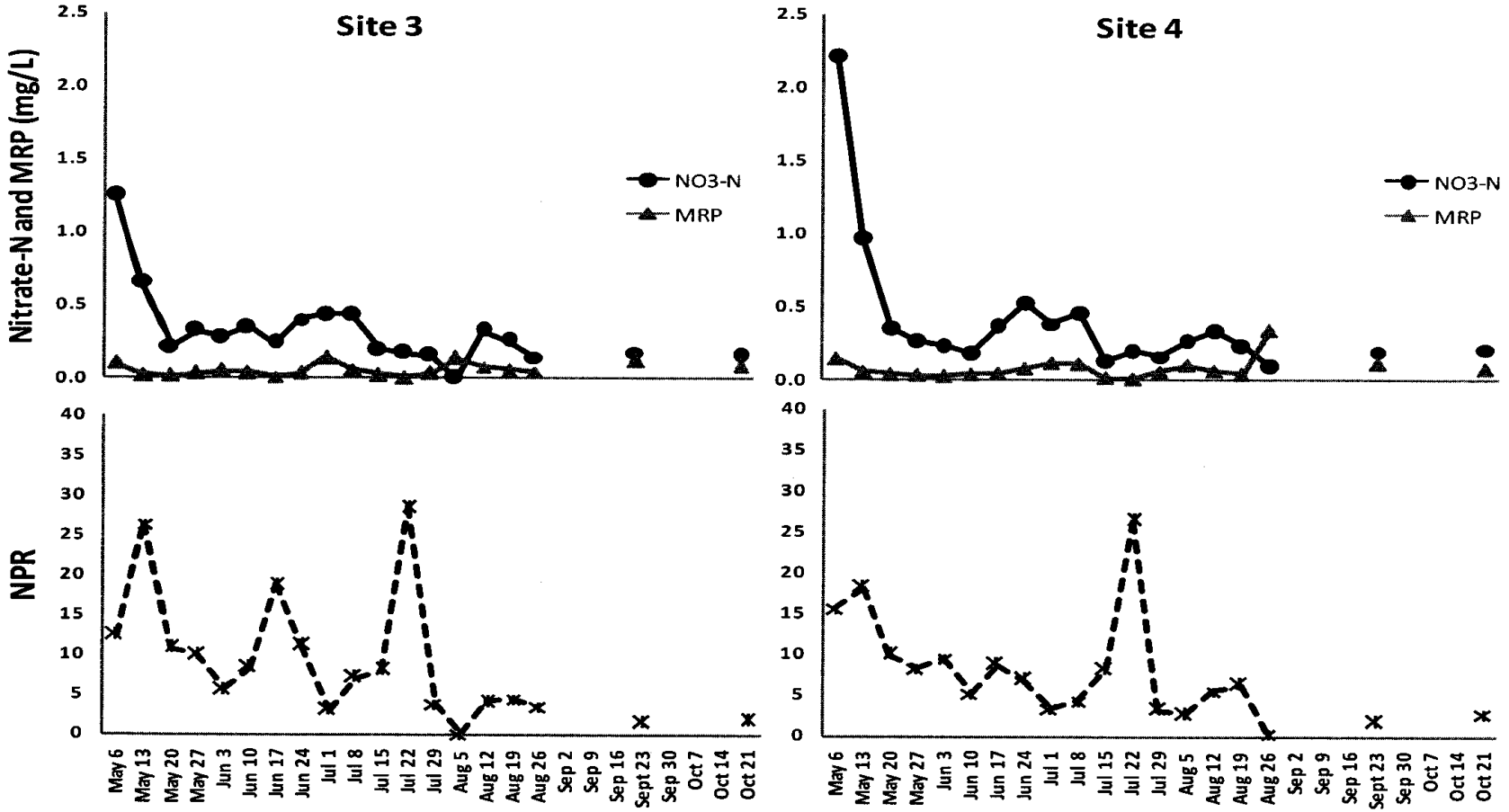
Appendix VIII: Seasonal fluctuations in chloride at four sites on the eastern shore of Lake Winnipeh during the 2007 ice-free season.



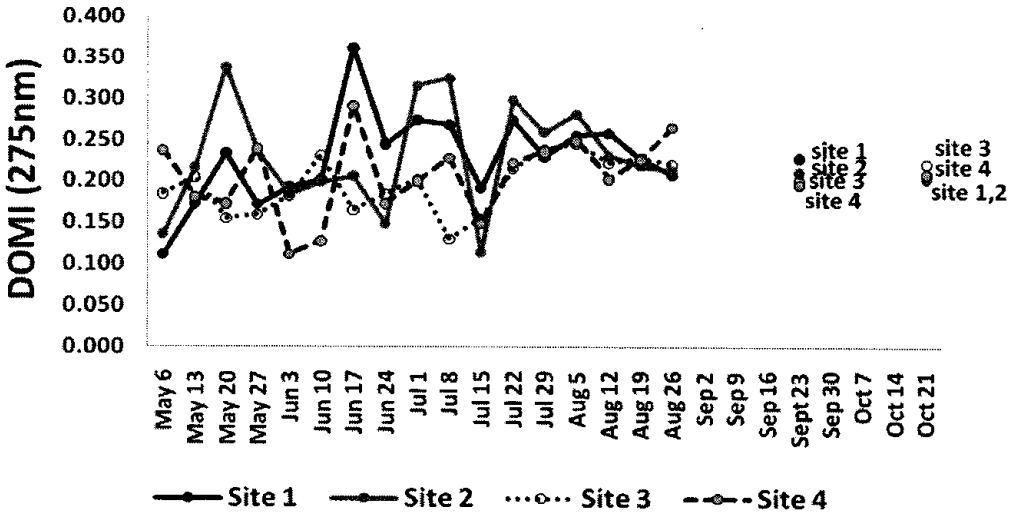
Appendix IX: Seasonal fluctuations in sulphate at four site on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



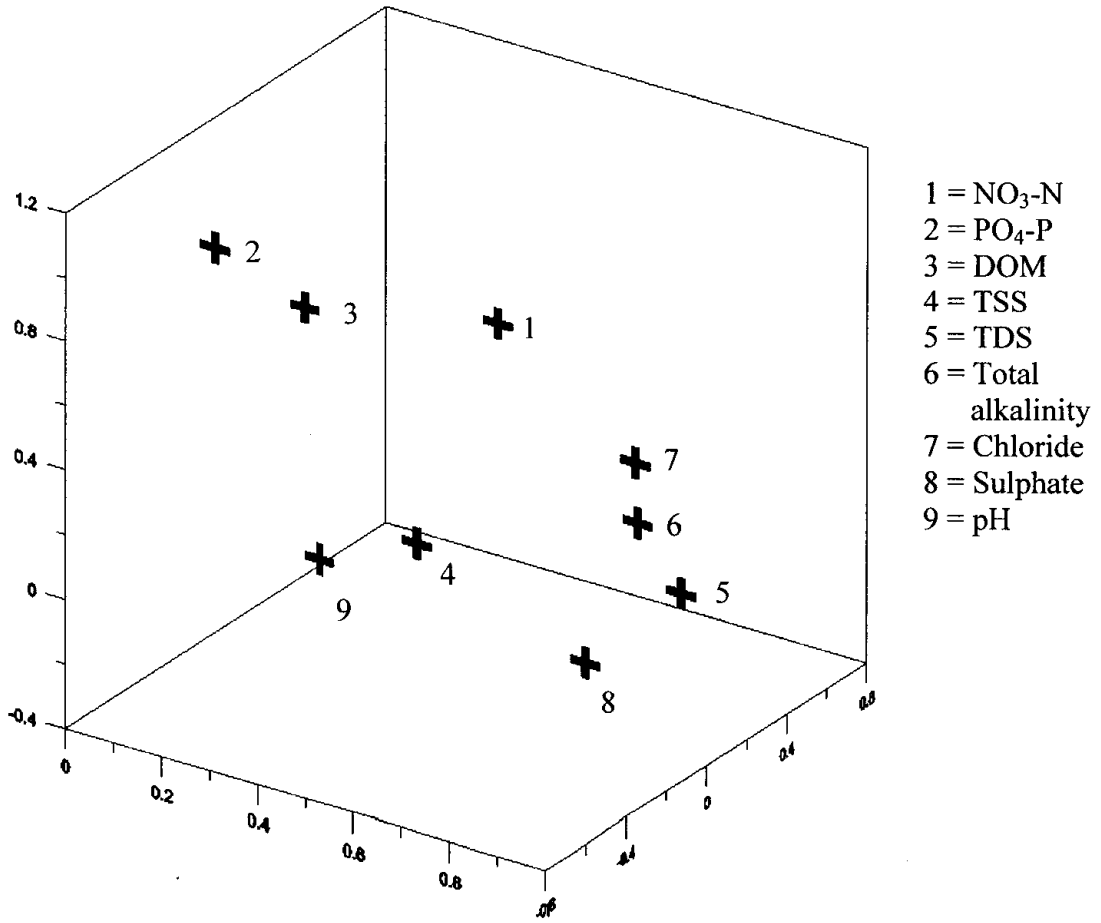
Appendix X-A: Seasonal fluctuations of nitrate-N, MRP, and NPR at Sites 1 and 2 on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



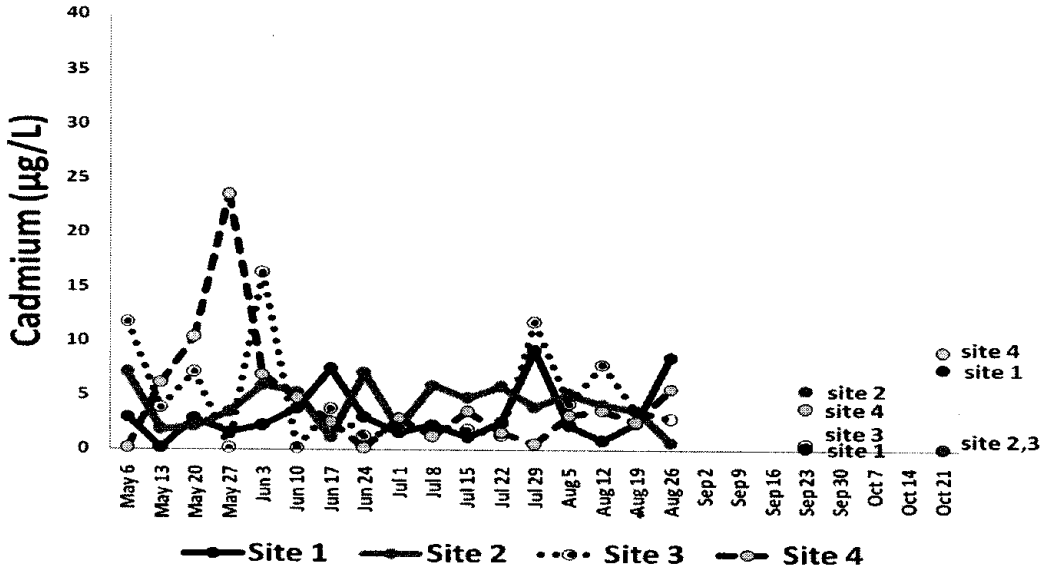
Appendix X-B: Seasonal fluctuations of nitrate-N, MRP, and NPR at Sites 3 and 4 on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



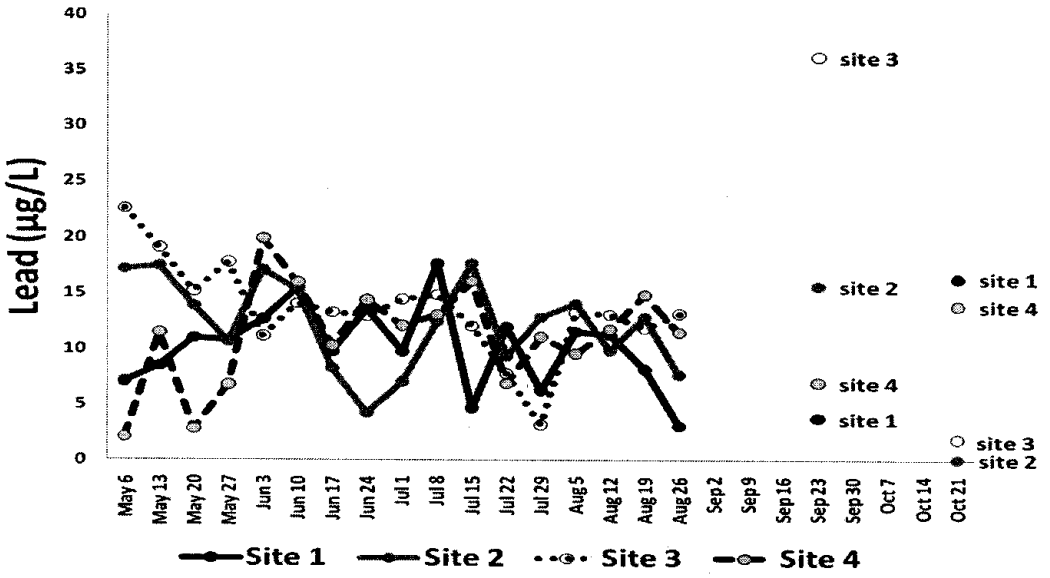
Appendix XI: Seasonal fluctuations in DOMI at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



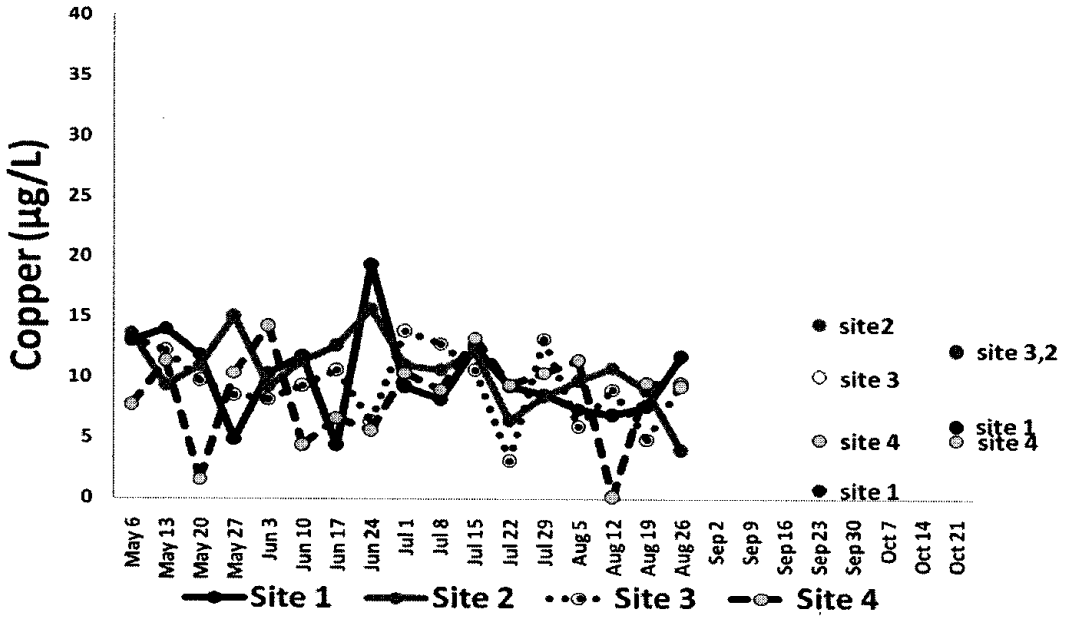
Appendix XII: Similarities of chemical parameters in terms of the first three rotated principle components.



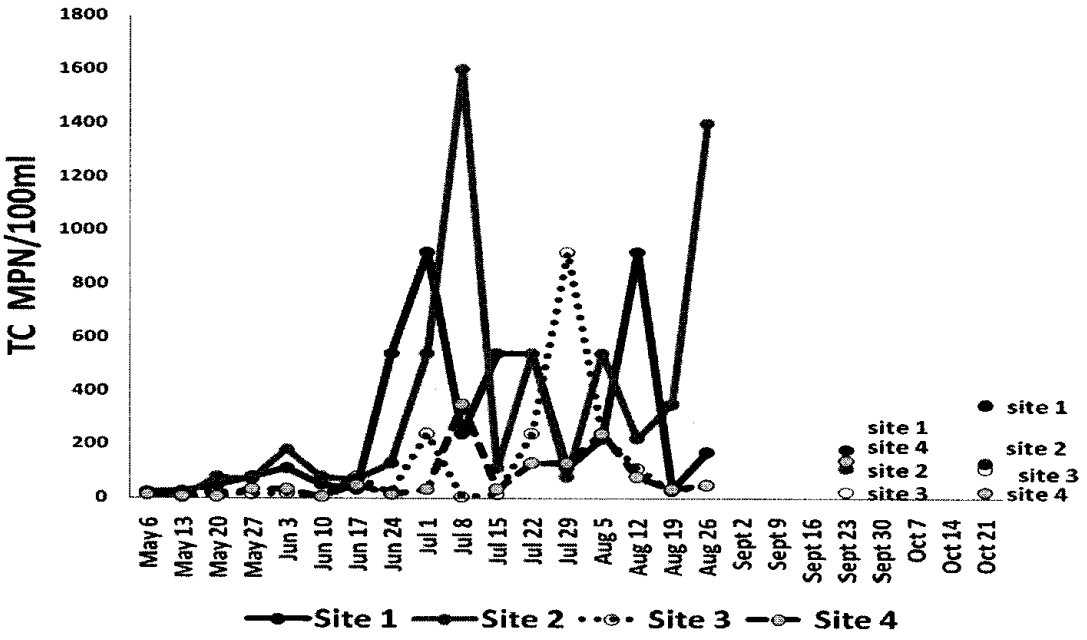
Appendix XIII: Seasonal fluctuations of cadmium at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



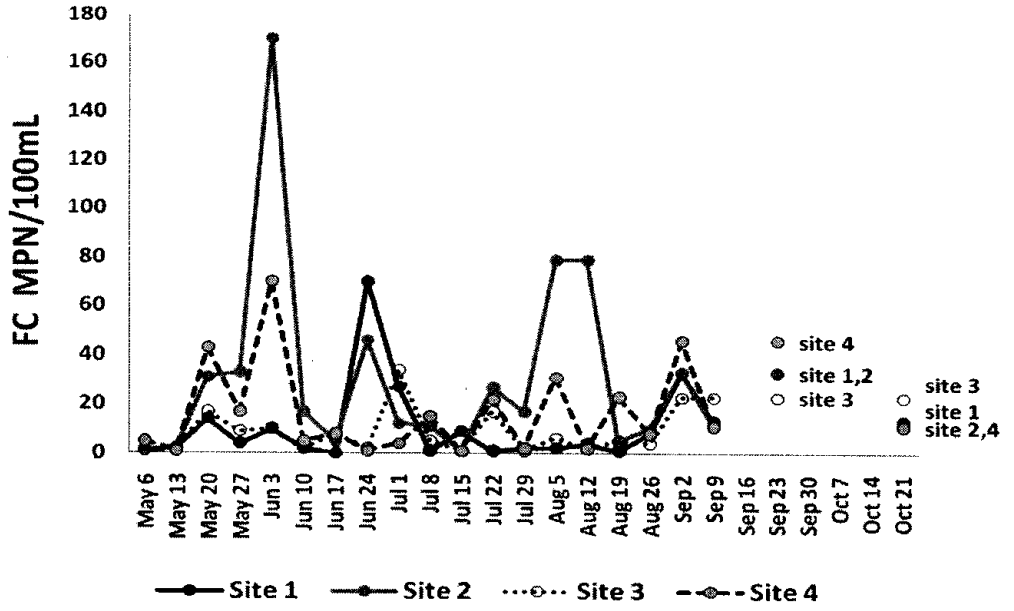
Appendix XIV: Seasonal fluctuations of lead at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



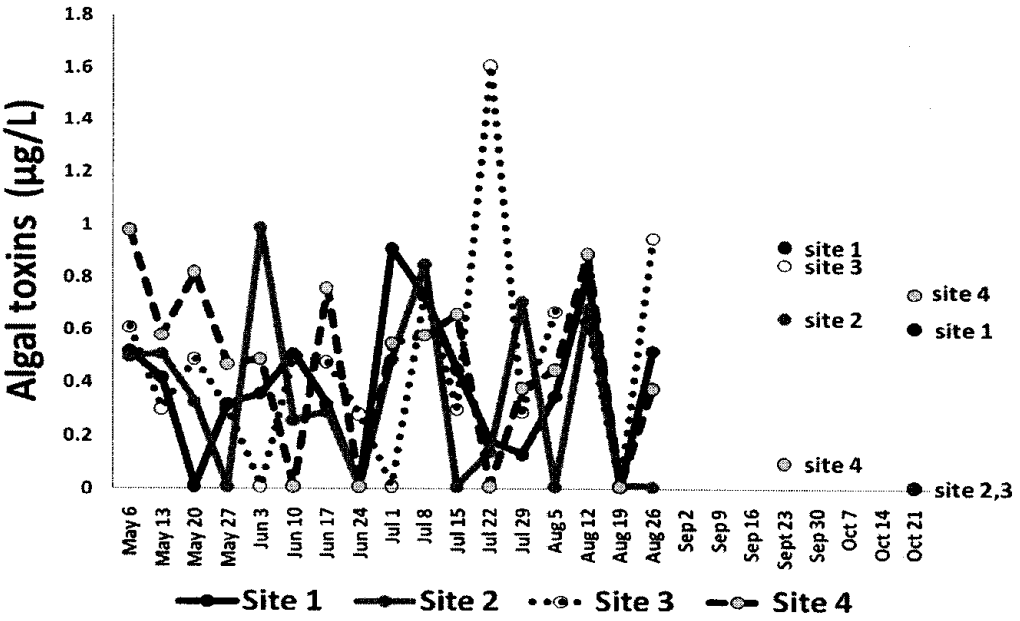
Appendix XV: Seasonal fluctuations of copper at four sites on the eastern shore of Lake Winnipeg during the ice-free 2007 season



Appendix XVI: Seasonal fluctuations in total coliform counts at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



Appendix XVII: Seasonal fluctuations in fecal colifom counts at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.



Appendix XVIII: Seasonal fluctuations in cyanobacterial toxins at four sites on the eastern shore of Lake Winnipeg during the 2007 ice-free season.