Assessment of *E. coli* and Microcystins in *Cladophora* Mats in the Nearshore Waters of Grand Traverse Bay, Little Traverse Bay, and Saginaw Bay

Prepared By

Richard R. Rediske, Ph.D., James O'Keefe, Kate Rieger, and Jeremy D. Rediske

> Annis Water Resources Institute Grand Valley State University 740 West Shoreline Drive Muskegon, Michigan 49441

Prepared For

Michigan Department of Environmental Quality 481062-07

May 2010

TABLE OF CONTENTS

TABLE OF CONTENTS	i
LIST OF TABLES	
LIST OF FIGURES	iii
Acknowledgements	iv
Executive Summary	v
1.0 Introduction	1
2.0 Methods	2
2.1 Sampling methods	2
2.1.1. Sampling Design	2
2.1.2. Sampling Methods	2
2.2 Analytical Methods	
2.2.1. Hydrolab Methods	5
2.2.2. E. coli Analysis	5
2.2.3. Sample Preparation for Microcystin Analysis	6
2.4.4. Microcystin LR and RR by LC/MS.	6
3.0 Results	
3.1 Saginaw Bay <i>E coli</i> , Microcystin, and Water Chemistry Results	6
3.1.1. Wenona Beach	
3.1.2. Bay City Recreation Area	
3.1.3. Brissette Beach.	
3.1.4. South Linwood Beach	
3.1.5. Pinconning Park	
3.1.6. White's Beach	
3.1.7. Arenac County Park	
3.1.8. Foster Road Beach	23
3.2 Grand Traverse Bay and Little Traverse Bay <i>E coli</i> , Microcystin, and Water	
Chemistry Results	
3.2.1. Petoskey State Park	
3.2.2. Magnus Park	
3.2.3. Elk Rapids Park	
3.2.4. Antrim County Day Park	
3.2.5. East Bay Park	
3.2.6. Traverse City State Park	
3.2.7. Bryant Park	
3.2.8. Clinch Park	
3.2.9. West End Beach	
3.3 Saginaw Bay and Grand Traverse Bay/Little Traverse Bay Comparisons	
4.0 Discussion	
5.0 Conclusions	
60 References	49

LIST OF TABLES

Table 2. 1. Laboratory Analytical Methods	5
Table 3.1.1. Wenona Beach Water Quality Data 2008	8
Table 3.1.2. Wenona Beach Cladophora Data 2008.	
Table 3.1.3. Bay City Recreation Area Water Quality Data 2008	
Table 3.1.4. Bay City Recreation Area <i>Cladophora</i> Data 2008	
Table 3.1.5. Brissette Beach Water Quality Data 2008.	
Table 3.1.6. Brissette Beach <i>Cladophora</i> Data 2008	
Table 3.1.7. South Linwood Beach Water Quality Data 2008.	
Table 3.1.8. South Linwood Beach <i>Cladophora</i> Data 2008.	
Table 3.1.9. Pinconning Park Water Quality Data 2008	
Table 3.1.10. Pinconning Park Cladophora Data 2008.	
Table 3.1.11. White's Beach Water Quality Data 2008.	
Table 3.1.12. White's Beach <i>Cladophora</i> Data 2008.	
Table 3.1.13. Arenac County Park Water Quality Data 2008.	
Table 3.1.14. Arenac County Park Cladophora Data 2008.	
Table 3.1.15. Foster Road Beach Water Quality Data 2008	
Table 3.1.16. Foster Road Beach Cladophora Data 2008.	
Table 3.2.1. Petoskey State Park Water Quality Data 2008	27
Table 3.2.2. Petoskey State Park Cladophora Data 2008	
Table 3.2.3. Magnus Park Water Quality Data 2008.	
Table 3.2.4. Magnus Park Cladophora Data 2008.	29
Table 3.2.5. Elk Rapids Park Water Quality Data 2008.	31
Table 3.2.6. Elk Rapids Park Cladophora Data 2008	
Table 3.2.7. Antrim County Day Park Water Quality Data 2008	
Table 3.2.8. Antrim County Day Park Cladophora Data 2008.	
Table 3.2.9. East Bay Park Water Quality Data 2008.	
Table 3.2.10. East Bay Park Cladophora Data 2008.	36
Table 3.2.11. Traverse City State Park Water Quality Data 2008.	38
Table 3.2.12. Traverse City State Park Cladophora Data 2008.	38
Table 3.2.13. Bryant Park Water Quality Data 2008.	
Table 3.2.14. Bryant Park Cladophora Data 2008.	
Table 3.2.15. Clinch Park Water Quality Data 2008.	42
Table 3.2.16. Clinch Park Cladophora Data 2008	
Table 3.2.17. West End Beach Water Quality Data 2008.	45
Table 3.2.18. West End Beach Cladophora Data 2008.	
Table 3.3.1. Summary of Water Quality and <i>Cladophora</i> Data for Grand Traverse	e Bay/Little
Traverse Bay and Saginaw Bay 2008.	

LIST OF FIGURES

Figure 2.1. Grand Traverse Bay/Little Traverse Bay Sampling Locations.	3
Figure 2.2. Saginaw Bay Sampling Locations.	
Figure 2.1.1 Wenne Book Sampling Locations 2009 (I = Loft C=Center D=Dight)	7
Figure 3.1.1. Wenona Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)	
Figure 3.1.2. E. coli in Detached Cladophora and Water at Wenona Beach 2008.	9
Figure 3.1.3. Bay City Recreation Area Sampling Locations 2008. (L= Left, C=Center, R=Right)	10
6 /	10
Figure 3.1.4. <i>E. coli</i> in Detached <i>Cladophora</i> and Water at the Bay City Recreation Area 2008	11
Figure 3.1.5. Brissette Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)	
Figure 3.1.6. E. coli in Detached Cladophora and Water at Brissette Beach 2008	
Figure 3.1.7. South Linwood Beach Sampling Locations 2008. (L= Left, C=Center,	13
R=Right)	11
Figure 3.1.8. <i>E. coli</i> in Detached <i>Cladophora</i> and Water at South Linwood Beach 2008	
Figure 3.1.9. Pinconning Park Sampling Locations 2008. (L= Left, C=Center, R=Right)	
Figure 3.1.10. E. coli in Detached Cladophora and Water at Pinconning Park 2008	
Figure 3.1.11. Whites Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)	
Figure 3.1.12. E. coli in Detached Cladophora and Water at White's Beach 2008.	
	20
Figure 3.1.13. Arenac County Park Sampling Locations 2008. (L= Left, C=Center, R=Right)	21
R=Right)	
Figure 3.1.15. Foster Road Beach Sampling Locations 2008. (L= Left, C=Center, R=Right) Figure 3.1.16. <i>E. coli</i> in Detached <i>Cladophora</i> and Water at Foster Road Beach 2008	
rigule 3.1.10. E. con in Detached Chaophora and water at Foster Road Beach 2008	23
Figure 3.2.1. Petoskey State Park Sampling Locations 2008. (L= Left, C=Center, R=	
Right)	26
Figure 3.2.2. E. coli in Detached Cladophora and Water at Petoskey State Park 2008	
Figure 3.2.3. Magnus Park Sampling Locations 2008. (L= Left, C=Center, R=Right)	
Figure 3.2.4. E. coli in Detached Cladophora and Water at Magnus Park 2008.	
Figure 3.2.5. Elk Rapids Park Sampling Locations 2008. (L= Left, C=Center, R=Right)	
Figure 3.2.6. E. coli in Detached Cladophora and Water at the Elk Rapids Park 2008	
Figure 3.2.7. Antrim County Day Park Sampling Locations 2008. (L= Left, C=Center,	
R=Right)	32
Figure 3.2.8. E. coli in Detached Cladophora and Water at the Antrim County Day Park	
2008	34
Figure 3.2.9. East Bay Park Sampling Locations 2008. (L= Left, C=Center, R=Right)	35
Figure 3.2.10. E. coli in Detached Cladophora and Water at East Bay Park 2008.	
Figure 3.2.11. Grand Traverse State Park Sampling Locations 2008. (L= Left, C=Center,	
R=Right)	37
Figure 3.2.12. <i>E. coli</i> in Detached <i>Cladophora</i> and Water at Grand Traverse State Park	- '
2008	38
Figure 3.2.13. Bryant Park Sampling Locations 2008. (L= Left, C=Center, R=Right)	39
Figure 3.2.14. E. coli in Detached Cladophora and Water at Bryant Park 2008.	
Figure 3.2.15. Clinch Park Sampling Locations 2008. (L= Left, C=Center, R=Right)	

Figure 3.2.16.	E. coli in Detached Cladophora and Water at Clinch Park 2008	43
Figure 3.2.17.	West End Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)	44
Figure 3.2.18.	E. coli in Detached Cladophora and Water at West End Beach 2008.	45

Acknowledgements

We thank the Michigan Department of Natural Resources and Environment for awarding AWRI a Clean Michigan Initiative - Clean Water Fund grant (TC: 481062-07), which funded this research and for the contributions of Tamara Lipsey as project Administrator.

We thank the AWRI staff that assisted with this project. Brian Scull assisted with the water chemistry analyses, and Jen Gradisher and Anthony Straley were involved with the preparation of cyanobacteria samples. For the work in the Grand Traverse Bay area, we thank Hans VanSummeren and the Water Studies Institute at Northwestern Michigan College for providing laboratory space for the analysis of *Cladophora* and *E. coli* samples and for coordinating outreach activities. We also thank Sara Uren of the Watershed Center Grand Traverse Bay for her assistance in the coordination of the area beach sampling program and the Inland Seas Education Association and the Tip of the Mitt Watershed Association for assistance with outreach activities. For the work conducted in Saginaw Bay, we thank Robert Hill of the Bay County Health Department for his assistance in site location and the coordination of the area beach monitoring program. We also thank Charlie Bauer of the MDNRE and the Bay City State Recreation Area for coordinating and hosting outreach activities.

Executive Summary

An assessment of the ability of *Cladophora* mats to sequester *E. coli* and microcystin LR and RR was conducted in the near-shore waters of Grand Traverse Bay (7 sites), Little Traverse Bay (2 sites), and Saginaw Bay (8 sites). The sampling locations were at public beach access points where *Cladophora* mats were previously observed. The goals of this research were to determine the spatial and temporal variability of *E. coli* populations in *Cladophora* mats in these recreational waters and if cyanotoxins (microcystin LR and RR) are sequestered in the detached algae. The collection of *Cladophora* samples was coordinated with local beach monitoring programs to facilitate the comparison with ambient water bacteria concentrations. This project provided important data for the assessment of public health impacts and the development of beach management programs to address the problems associated with *Cladophora* accumulations.

Based on the results from this investigation, Saginaw Bay appears to be more heavily impacted by detached Cladophora than Grand Traverse/Little Traverse Bays. Mean E. coli concentrations in detached Cladophora were higher in Saginaw Bay (2,796 cfu/g dwt) than Grand Traverse Bay/Little Traverse Bay (1,775cfu/g dwt); however, the difference was not statistically significant (Mann-Whitney p=0.40). Cladophora deposits exhibited spatial and temporal variability in both systems. At most beaches in Grand Traverse Bay, Cladophora deposits were limited to small pockets at 1 location. Clinch Park had only one site with Cladophora on the last sampling event and two locations at the Traverse City State Park were free of detached algal accumulations. In contrast, Cladophora deposits in Saginaw Bay covered approximately 1 meter (m) of the shoreline at most beaches. Two locations in Saginaw Bay also had no accumulations of Cladophora during the study period (White's Beach and Pinconning Park). Differences in Cladophora accumulation between Saginaw Bay and Grand Traverse Bay/Little Traverse Bay may be attributed to higher total phosphorus levels in Saginaw Bay. Levels of E. coli in detached *Cladophora* in both systems were similar to concentrations previously reported in the Great Lakes (1,000 cfu/g dwt - 60,000 cfu/g dwt). In Saginaw Bay, the highest levels of E. coli in detached Cladophora were consistently found at beaches near the Saginaw River. Even within individual sites, locations near tributaries and drains at Wenona Beach and South Linwood Beach were significantly higher than locations farther away from a point source. This relationship also was noted in Grand Traverse Bay, where the location near Mitchell Creek at the Traverse City State Park, had elevated E. coli concentrations in detached Cladophora compared to the other beach locations. These results suggest that *Cladophora* can trap bacteria from point sources and also be stimulated by nutrient discharges. T wo locations, Pinconning Park and White's Beach, had very limited Cladophora growth. Both locations had Chara growing on the lake bottom. Chara is known to exhibit allelopathic activity that can limit the growth of other aquatic plants. No correlation was found between E. coli levels in the open water (designated beach monitoring locations) and the near-shore zone, where the detached Cladophora samples were taken. As noted in previous studies, Cladophora appears to hold trapped E. coli and does not release the entrained bacteria into the offshore water.

This investigation was the first to document the accumulation of microcystins in the detached *Cladophora* of Saginaw Bay. Total microcystins in detached *Cladophora* had a grand mean of 57 µg/g dwt for the study period. Saginaw Bay has a history of *Microcystis* blooms in the late

summer months that produce both microcystin LR and RR. Since *Microcystis* has a high requirement for sunlight, cyanobacteria may become stressed when they are trapped in the detached algae mats. While accidental ingestion by humans of microcystins trapped in *Cladophora* is unlikely, these compounds can act as skin irritants. Walking through *Cladophora* accumulations to get to deeper water may provide sufficient exposure to cause irritation in sensitive individuals if microcystins are present. Although the data suggest that swimming areas (1 m depth) are not impacted by the *E. coli* accumulations in detached *Cladophora*, entrained bacteria and cyanotoxins may pose a hazard to children playing in the nearshore water and beach sand. Current regulations discourage beach grooming and altering the nearshore zone. The presence of elevated bacteria and microcystin levels in the nearshore environment of Saginaw Bay suggests that the current policy should be reevaluated to balance potential impacts to public health with the ecosystem services provided by coastal wetlands.

1.0 Introduction

Cladophora is a filamentous green alga that commonly grows attached to hard substrates in the littoral zone of freshwater environments and is widely distributed throughout the Laurentian Great Lakes (Graham 1982, Dodds and Gudder 1992). This macroalga is considered a public nuisance, causing shoreline fouling and clogging of water intake pipes (Joska and Bolton 1996). When mats of Cladophora detach and wash onshore, their subsequent decay is associated with notoriously foul odors (Higgins et al. 2005; Wilson et al. 2006), nearshore oxygen depletion (Whitman et al. 2003), and elevated bacterial loads (Byappanahalli 2003). In the Laurentian Great Lakes, Cladophora is a nuisance throughout Lakes Ontario, Erie, Huron, and Michigan, where hard substrates are available (Higgins et al. 2005). Public concern over detached and decaying Cladophora along beaches in the lower Great Lakes began as early as the 1950s-1960s, and peaked in the 1970s (Bartsch 1968; Taft 1975). Shoreline fouling by decaying macroalgae was a significant factor for limiting nutrient loading to the Great Lakes, as identified in the amended Great Lakes Water Quality Agreement (GLWQA) of 1978. Subsequent phosphorus management programs led to a decrease in Cladophora biomass in the early 1980s (Painter and Kamaitis 1987). Beginning in the late 1990s, the dominance of invasive dreissenid mussels (i.e. the zebra mussel *Dreissena polymorpha* and the quagga mussel *D. bugensis*) in the Laurentian Great Lakes has reengineered nutrient distributions by removing suspended particulate matter through filter feeding and generating heavier particulate matter as feces and pseudofeces in the benthos (Hecky et al. 2004). This process has been implicated in shifting pools of organic matter and nutrients from the pelagic zone to the benthos and increasing the solar radiant flux to the Since the invasion of dreissenids, Cladophora biomass has benthos (Zhu et al. 2006). dramatically increased in the lower Great Lakes (Mills et al. 2003; Bootsma et al. 2005; Hecky et al. 2004).

The occurrence of Cladophora in near-shore Lake Michigan habitats may have significant public health and ecological importance (Whitman et al. 2003), and could potentially lead to a decrease in recreational activities on beaches. The presence of Cladophora has been associated with high levels of E. coli and other species of enteric bacteria in the beach sand and swimming waters of the Great Lakes (Whitman et al. 2003; Olapade et al. 2006). These bacteria may even grow on Cladophora under certain conditions (Byappanahalli et al. 2003) and the alga can potentially harbor and enhance the survival of pathogenic bacteria released into the environment through point and non-point sources. Cladophora has been reported to provide nutrients and protect attached bacteria from environmental stresses, such as desiccation, predation, and harmful radiation (Byappanahalli et al. 2003; Englebert et al. 2008). Ishii et al. (2006) found several enteric bacterial pathogens, including Campylobacter, shiga toxin-producing E. coli (STEC), Shigella, and Salmonella, from Cladophora at a southern Lake Michigan beach. In addition to pathogens, sulfate-reducing bacteria and other phylogenetically diverse groups of microorganisms also have been detected in *Cladophora* mats (Olapade et al. 2006). Recently, Clostridium botulinum and Clostridium perfringens were found to grow in Cladophora mats (Byappanahalli and Whitman 2009). In this manner, Cladophora mats may play a significant role as a source and sink for pathogens in the near-shore environment. Concerns related to Cladophora mats and their ability to sequester pathogens have been raised in Michigan as part of watershed plans for Grand Traverse Bay, Little Traverse Bay, and Saginaw Bay. In addition, the Saginaw Bay Area of Concern has Beach Closings listed as a Beneficial Use Impairment due to elevated E. coli levels.

Although the public health and ecological importance of microbial communities associated with *Cladophora* mats in freshwater environments have been previously studied, their role in sequestering potentially toxic cyanobacteria is poorly understood. Nuisance blooms of *Microcystis* have been reported in Saginaw Bay (Vanderploeg et al. 2001; Fahnensteil et al, 2008) and several of the large inland lakes near Grand Traverse Bay (Keilty and Woller 2004). Since cyanobacteria blooms also can accumulate on beaches and cause fouling of the shoreline (Codd 1995; Landsberg 2002), the presence of cyanotoxins in detached *Cladophora* mats should be investigated.

We conducted an assessment of the ability of *Cladophora* mats to sequester *E. coli* and microcystin LR and RR in the near-shore waters of Grand Traverse Bay (7 sites), Little Traverse Bay (2 sites), and Saginaw Bay (8 sites). The sampling locations were at public beach access points where *Cladophora* mats have been previously observed. The goals of this research were to determine the spatial and temporal variability of *E. coli* populations in *Cladophora* mats in these recreational waters, and if cyanotoxins (microcystin LR and RR) are sequestered in the detached algae. The collection of *Cladophora* samples was coordinated with local beach monitoring programs to facilitate the comparison with ambient water bacteria concentrations. This project provided important data for the assessment of public health impacts and the development of beach management programs to address the problems associated with *Cladophora* accumulations.

2.0 Methods

2.1 Sampling methods

2.1.1. Sampling Design

Eight locations in Saginaw Bay, seven locations in Grand Traverse Bay, and two locations in Little Traverse Bay were sampled in triplicate for *E. coli* and microcystin LR and RR during the summer of 2008. The locations were sampled weekly for 8 weeks in June-August. Grab samples of floating *Cladophora* mats were collected at each location and analyzed individually for *E. coli* and as a composite for microcystin LR and RR. Sampling dates and times were coordinated with local beach monitoring programs to facilitate the use of ambient water bacteria concentrations collected close to the time of *Cladophora* sampling. *Cladophora* was collected and prepared as described by Whitman et al. (2003). The use of standardized collection protocols and triplicate samples provided a high level of quality assurance and resulted in data that were appropriate for decision making and environmental assessment.

2.1.2. Sampling Methods

Triplicate *Cladophora* samples were collected at each site with an integrated collection device at 0.25-0.5 m depth (Sutherland et al. 1992). The sampling locations for Grand Traverse Bay/Little Traverse Bay and Saginaw Bay are shown in Figures 1 and 2, respectively. Each location was marked with GPS coordinates and sampled at weekly intervals during June - August 2008 (8)

sampling events). *E. coli s*amples were placed in sterile whirl pack bags and stored in the dark at 4°C. A composite of the triplicate samples was placed in an amber glass jar and stored at 4°C for microcystin-LR and RR analysis. All samples were returned to the laboratory and on a daily basis for further processing and storage. *E. coli* samples were analyzed within 6 hours of collection. Microcystin –LR and RR samples were lyophilized and stored at -30°C.

Chemical and physical parameters were monitored at each location using a Hydrolab DataSonde 4a. We measured pH, dissolved oxygen (DO), DO% saturation, temperature, and specific conductance at each station.

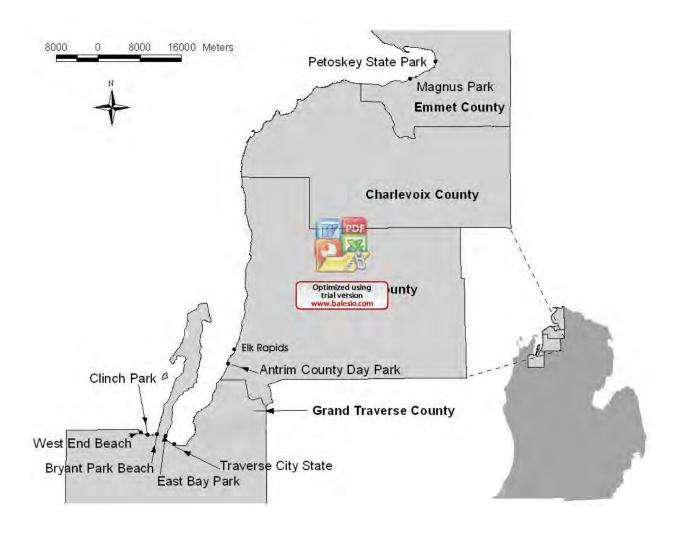


Figure 2.1. Grand Traverse Bay/Little Traverse Bay Sampling Locations.

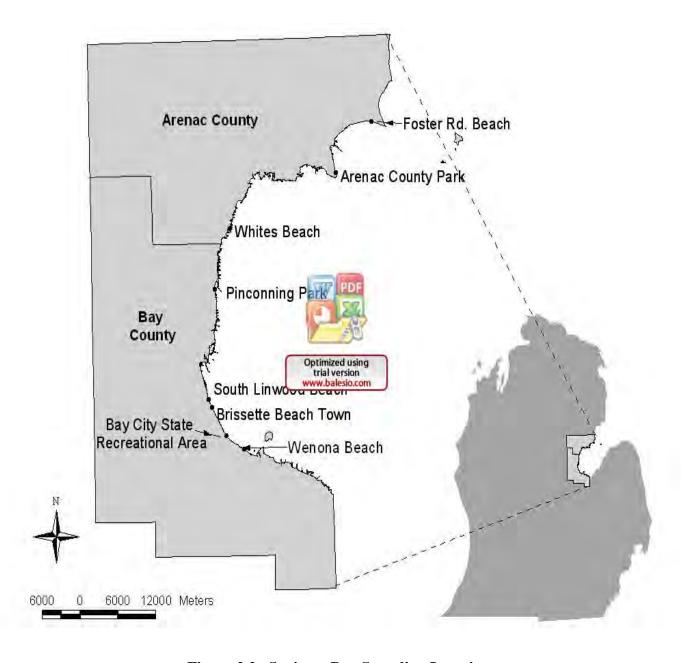


Figure 2.2. Saginaw Bay Sampling Locations.

The extent of *Cladophora* accumulation was determined at each sampling location according to the ranking:

- 0 = No visible Cladophora
- 1 = *Cladophora* present in isolated pockets at one location
- 2 = Cladophora present in isolated pockets at all locations
- 3 = Cladophora residue 1 m² in area
- 4 = Cladophora residue 1-5 m² in area
- $5 = Cladophora > 5 \text{ m}^2 \text{ in area}$

2.2. Analytical Methods

Laboratory procedures have been selected based on previous use for water quality investigations, established ranges for accuracy/precision, and limited problems related to matrix interferences. A summary of analytical methods is given in Table 2.1. Method summaries are provided in the following sections.

2.2.1. Hydrolab Methods

Hydrolab measurements of pH, temperature, dissolved oxygen, and specific conductance were conducted according to the methods in the instrument users manual (Hydrolab 1998). The Hydrolab Datasonde 4a was calibrated in the lab prior to use in the field. The instrument was placed in the water at each site and the technician waited for a stable reading of the sensors. Measurements were recorded at 0.5 m depth at each station. Precision was assessed by the measurement of field duplicates at 10% of the stations.

Table 2.1. Laboratory Analytical Methods

Parameter	Preparation	Preservation	Holding Time	Methods Reference
pH*	*	*	*	Hydrolab 1998
Dissolved Oxygen*	*	*	*	Hydrolab 1998
Temperature*	*	*	*	Hydrolab 1998
Specific conductance*	*	*	*	Hydrolab 1998
Microcystin LR and RR by LC/MS	Filtration, methanol extraction, C-18 cartridge	Freeze -30°C	6 mo	Lawerence et al. 2001; Boyer et al. 2004
E. coli	Phosphate buffer extraction	Cool 4°C	6 hrs	Whitman et al. 2003 Colilert-18 APHA 1998

2.2.2. E. coli Analysis

Samples of *E. coli* were prepared by elutriation with phosphate buffer (Whitman et al. 2003). One gram portions of homogenized algal samples were weighed and placed in sterile 15-milliliter (mL) centrifuge tubes. Nine mL of sterile phosphate-buffered diluent water (pH 6.8) was added and the mixture was vigorously shaken for 2 min and then centrifuged briefly (45 s) at 2,000 rpm to allow the large particles to settle. The diluted sample then was analyzed by the Colilert-18 (IDEXX) using the Quanti-Tray 2000 (APHA 1998). A 1 mL aliquot was removed from the centrifuge tube and transferred to 99 mL of sterile phosphate buffer in a sterile water sampling vial. The Colilert reagent then was then added to the vial and shaken until dissolved. The sample and reagents were then added to the Quanti-Tray 2000 and sealed with the Quanti-Tray sealer. The tray was incubated for 18 ho urs at 35°C \pm 0.5°C and read under UV light. Fluorescent colonies were counted and the number of colony forming units (cfu)/100 mL was calculated according to the manufacturer's instructions.

2.2.3. Sample Preparation for Microcystin Analysis

Water samples for the analyses of microcystin were prepared according to methods outlined by Lawerence et al. (2001). A 100-500 mL aliquot was filtered through a Whatman GF/C glass fiber filter. The filter then was placed in a 5 mL centrifuge tube and lyophilized for 8 hours. The filter residue was extracted with three successive aliquots of methanol followed by sonication. The methanol extract was evaporated to dryness under nitrogen and re-dissolved in 1 mL of 50% methanol in water. This mixture was passed through a 1 gram (g) SPE C-18 cartridge (Baker) that had been preconditioned with 10 mL of methanol followed by 10 mL of water. The cartridge then was washed with 5 mL of 25% methanol in water and the eluted fraction was discarded. The microcystins then were eluted with 4 mL of 100% methanol in water. This fraction was collected and the volume reduced to 2 mL. The volume was adjusted back to 3 mL with methanol and split into three 1 mL aliquots for HPLC/MS analysis.

2.4.4. Microcystin LR and RR by LC/MS.

Samples for Liquid Chromatography/Mass Spectrometry (LC/MS) analysis were filtered using Gelman A/E binderless glass fiber filters. The cells were lysed by freezing and then vortexed for 5 minutes in centrifuge tubes containing 10 mL of aqueous (50:50) methanol to extract toxins. Suspensions were centrifuged and the supernatant was filtered through 0.45 micron nylon filters. The concentrations of microcystin LR and RR were determined by liquid chromatography-tandem mass spectrometry using a Finnigan Surveyor MSQ LC/MS. Nodularin was added to the extracts and used as an internal standard. Compounds were separated on a Phenomenex Luna C18 column at 50°C. The mobile phase was a binary gradient of water and methanol, both containing 0.1% formic acid. The initial gradient started with 95% water and 5% methanol, followed by a step change to 50% water and 50% methanol at 3 minutes, with a linear gradient from 5 to 20 minutes to 5% water and 95% methanol. Instrument detection limits for these toxins were determined to be 1 microgram/liter (μ g/l). For calibration, a series of 5 solutions were prepared with the internal standard at 1000 μ g/l and the analytes in the range of 1 to 500 μ g/l in final volumes of 1 mL of 90:10 water:methanol.

3.0 Results

The results of *E. coli*, water chemistry, and microcystins data from each location are presented for Saginaw Bay and Grand Traverse Bay/Little Traverse Bay in Sections 3.1 and 3.2, respectively. A comparison of the two basins is provided in Section 3.3.

3.1. Saginaw Bay *E. coli*, Microcystin, and Water Chemistry Results

3.1.1. Wenona Beach

The sampling locations for Wenona Beach are provided in Figure 3.1.1. Near-shore locations were sampled at a depth of 0.3 m. The beach area was covered with reed canary grass, sedges and cattails. Cattails and detached *Cladophora* were common in the shallow littoral zone and the samples were collected approximately 1 meter outside of the macrophyte zone. The detached

Cladophora also was common at the sampling locations with some areas of fresh *Cladophora* growing on submerged rocks and woody debris.



Figure 3.1.1. Wenona Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.1. and 3.1.2., respectively. The mean *E. coli* concentration for the study period was 14 cfu/100 mL with no single beach samples exceeding 100 cfu/100 mL (Table 3.1.1). Water temperatures ranged from 21°C to 25°C. The mean *E. coli* concentration for detached *Cladophora* was 9,775 cfu/g dwt (Table 3.1.2.). Microcystins were detected in the detached *Cladophora* beginning in July with mean concentrations of 47 μ g/g dwt, 47 μ g/g dwt, and 103 μ g/g dwt for microcystin RR, microcystin LR, and total microcystins, respectively. Detached *Cladophora* was present in

Table 3.1.1. Wenona Beach Water Quality Data 2008.

		E coli	in Water			Water	Chemistry	
Date		cfu/	100 ml		На	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	ρΠ	DO (IIIg/I)	remp C	μS/cm
06/23/08	10	8	18	12	7.92	7.7	21	440
07/01/08	78	44	82	68	8.10	7.8	22	470
07/08/08	3	4	2	3	8.14	7.4	23	451
07/15/08	2	1	1	1	8.17	7.5	24	450
07/22/08	5	6	1	4	8.16	7.3	24	456
07/29/08	33	8	7	16	8.10	7.2	25	453
08/05/08	4	7	4	5	8.13	7.6	24	461
08/12/08	4	2	3	3	8.11	7.9	24	467
Mean	17	10	15	14	8.1	7.6	23	456

Table 3.1.2. Wenona Beach Cladophora Data 2008.

Date	E.	. coli in Clad	dophora cfu/	g dwt	Cladophora	Microcysti	ns in <i>Cladophoi</i>	ra μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	448	733	22507	7896	5	< 0.1	< 0.1	< 0.1
07/01/08	800	2331	25432	9521	5	< 0.1	1	< 0.1
07/08/08	1279	3561	31566	12135	5	12	11	23
07/15/08	610	500	1678	929	3	15	23	38
07/22/08	1892	2000	3578	2490	5	27	34	61
07/29/08	5631	5633	64132	25132	5	68	80	148
08/05/08	6322	4531	25346	12066	5	80	80	161
08/12/08	3313	2311	18462	8029	5	82	102	184
Mean	2537	2700	24088	9775	5	47	47	103

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2$ in area; $4 = \text{Deposits } 1 - 5 \text{ m}^2$ in area; $5 = \text{Deposits } > 5 \text{ m}^2$ in area.)

deposits of >5 m² at all locations during the sampling period (Table 3.1.2.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.1.2. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.14). The drop in *E. coli* levels observed on July 15 was after a storm that had occurred on July 12. A statistically significant difference was observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.0.004). Station R was significantly different from C and L (Mann-Whitney ρ =0.012 and 0.010, respectively). Station R was the closest sampling location to the Saginaw River. Station R had the highest concentration of *E. coli* in detached *Cladophora* (64,132 cfu/g dwt). This bacterial concentration was the highest level observed during the study period.

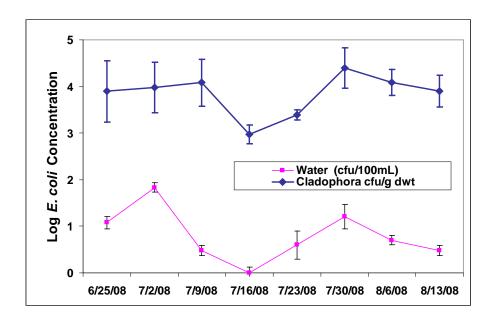


Figure 3.1.2. E. coli in Detached Cladophora and Water at Wenona Beach 2008.

3.1.2. Bay City Recreation Area

The sampling locations for the Bay City Recreation Area are provided in Figure 3.1.3. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be groomed. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1 meter from shore.

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.3. and 3.1.4., respectively. The mean E. coli concentration for the study period was 6 cfu/100 mL with no single beach sample exceeding 100 cfu/100 mL (Table 3.1.3.). Water temperatures ranged from 21°C to 24°C. The mean E. coli concentration in detached Cladophora was 3.216 cfu/g dwt (Table 3.1.4.). Microcystins were detected in the detached Cladophora beginning in June with mean concentrations of 96 μg/g dwt, 129 μg/g dwt, and 225 μg/g dwt for microcystin RR, microcystin LR, and total microcystins, respectively. The highest concentrations of microcystins were found on August 12 at the Bay City Recreation area (microcystin RR, 364 µg/g dwt; microcystin LR 349 µg/g dwt; and total microcystins 714 µg/g dwt). Detached Cladophora covered the entire beach for all but one of the sampling events. E. coli concentrations in water and detached *Cladophora* are displayed in Figure 3.1.4. No correlation was observed between bacterial levels in the water and the detached algae (Spearman's RO ρ =0.08). The drop in E. coli levels observed on July 15 was after a storm that had occurred on July 12. significant difference was not observed between beach locations for E. coli concentrations in detached Cladophora (Kruskal-Wallis ρ =0.0.073). Station R had the highest concentration of E. coli in detached Cladophora (8,923 cfu/g dwt).



Figure 3.1.3. Bay City Recreation Area Sampling Locations 2008. (L= Left, C=Center, R=Right)

Table 3.1.3. Bay City Recreation Area Water Quality Data 2008.

		<i>E coli</i> in	Water			Water	Chemistry	
Date			рН	DO (mg/l)	Temp ^O C	Conductance		
	L	С	R	Mean	рп	DO (IIIg/I)	remp C	μS/cm
06/23/08	10	8	24	14	7.92	7.7	21	420
07/01/08	1	2	6	3	8.11	7.8	22	437
07/08/08	3	4	47	18	8.16	7.5	23	445
07/15/08	2	1	1	1	8.16	7.4	23	436
07/22/08	1	1	1	1	8.16	7.3	24	436
07/29/08	4	8	1	4	8.12	7.7	24	446
08/05/08	4	7	4	5	8.17	7.8	24	448
08/12/08	1	2	1	1	8.11	8.1	24	449
Mean	3	4	11	6	8.11	7.7	23	440

Table 3.1.4. Bay City Recreation Area Cladophora Data 2008.

Date	E. 0	coli in Clado	<i>phora</i> cfu/g c	dwt	Cladophora	Microcystins in	n <i>Cladophora</i>	μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	1432	856	3456	1915	5	1	1	2
07/01/08	1333	963	2564	1620	5	2	7	9
07/08/08	4222	4563	8923	5903	5	14	33	47
07/15/08	1100	1245	1344	1230	3	32	49	81
07/22/08	1632	2000	7843	3825	5	65	99	164
07/29/08	998	5633	8564	5065	5	56	219	275
08/05/08	2331	4531	3564	3475	5	231	275	505
08/12/08	3223	2311	2564	2699	5	364	349	714
Mean	2034	2763	4853	3216	5	96	129	225

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } 5 \text{ m}^2 \text{ in area}$.)

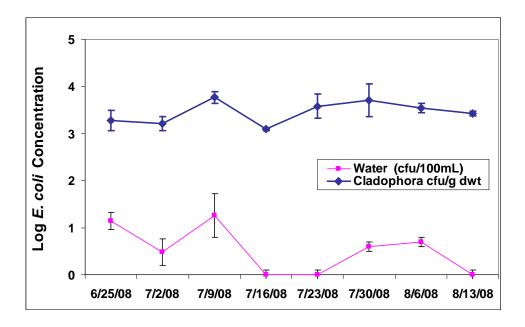


Figure 3.1.4. *E. coli* in Detached *Cladophora* and Water at the Bay City Recreation Area 2008.

3.1.3. Brissette Beach

The sampling locations for Brissette Beach are provided in Figure 3.1.5. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be groomed. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1-2 m from shore.



Figure 3.1.5. Brissette Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.5. and 3.1.6., respectively. The mean *E. coli* concentration for the study period was 7 cfu/100 mL with no single beach sample exceeding 100 cfu/100 mL (Table 3.1.5.). Water temperatures ranged from 21°C to 24°C. The mean *E. coli* concentration in detached *Cladophora* was 768 cfu/g dwt (Table 3.1.6.). Microcystins were detected in the detached *Cladophora* beginning in June with mean concentrations of 9 μ g/g dwt, 15 μ g/g dwt, and 23 μ g/g dwt for microcystin RR, microcystin LR, and total microcystins, respectively. Detached *Cladophora* was present in deposits of 1-5 m² at all locations during the sampling period except after the storm event on July 12 (Table 3.1.6.). *E. coli* concentrations in the water and detached *Cladophora* are displayed in Figure 3.1.6. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.83). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.0.53). Station R had the highest concentration of *E. coli* in detached *Cladophora* (1,848 cfu/g dwt).

Table 3.1.5. Brissette Beach Water Quality Data 2008.

		E coli ii	n Water			Water	Chemistry	
Date		cfu/1	00 ml		рН	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	рп	DO (IIIg/I)	remp C	μS/cm
06/23/08	3	2	1	2	7.89	7.7	21	400
07/01/08	15	22	35	24	8.12	7.8	22	440
07/08/08	1	1	1	1	8.19	7.5	23	433
07/15/08	2	1	3	2	8.12	7.4	23	434
07/22/08	1	1	7	3	8.12	7.3	24	444
07/29/08	16	37	16	23	8.18	7.7	24	439
08/05/08	1	2	1	1	8.19	7.8	24	445
08/12/08	1	2	6	3	8.13	8.1	24	447
Mean	5	9	9	7	8.12	7.7	23	435

Table 3.1.6. Brissette Beach Cladophora Data 2008.

Date	E.	. coli in Cladophora	a cfu/g dwt		Cladophora	Microcystin	s in <i>Cladopho</i>	ra μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	618	1045	1848	1170	4	0.5	0.7	1
07/01/08	450	533	460	481	4	0.1	0.7	1
07/08/08	655	784	989	809	4	2.3	3.7	6
07/15/08	411	333	450	398	2	0.9	7.7	9
07/22/08	644	755	877	759	4	3.5	13.1	17
07/29/08	755	766	864	795	4	1.8	14.2	16
08/05/08	644	532	436	537	4	21.3	28.5	50
08/12/08	1033	1200	1344	1192	4	38.8	49.4	88
Mean	651	744	908	768	4	9	15	23

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2$ in area; $4 = \text{Deposits } 1 - 5 \text{ m}^2$ in area; $5 = \text{Deposits } 5 \text{ m}^2$ in area.)

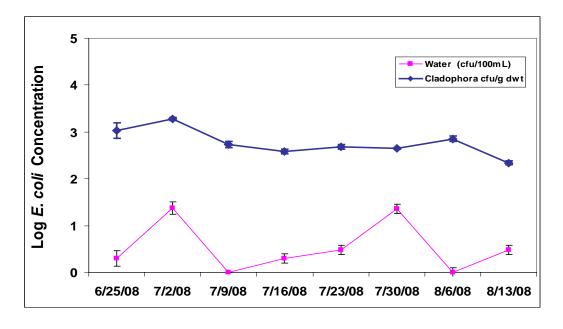


Figure 3.1.6. E. coli in Detached Cladophora and Water at Brissette Beach 2008.

3.1.4. South Linwood Beach

The sampling locations for South Linwood Beach are provided in Figure 3.1.7. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be groomed. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1-2 m from shore.

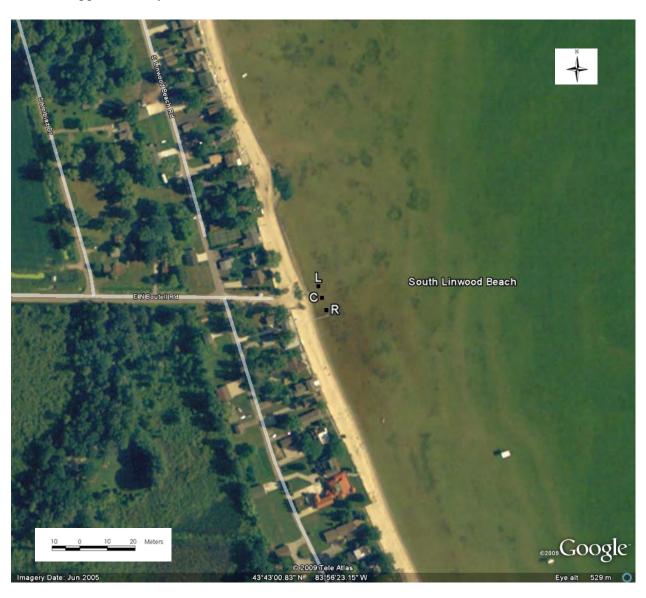


Figure 3.1.7. South Linwood Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.7. and 3.1.8., respectively. The mean E. coli concentration for the study period was 9 cfu/100 mL with no single beach sample exceeding 100 cfu/100 mL (Table 3.1.7.). Water temperatures ranged from 21°C to 24°C. The mean E. coli concentration in detached Cladophora was 1,191 c fu/g dwt (Table 3.1.8.). Microcystins were detected in the detached Cladophora beginning in July with mean concentrations of 12 μ g/g dwt, 15 μ g/g dwt, and 27 μ g/g dwt for microcystin RR, microcystin LR, and total microcystins, respectively. Detached Cladophora was present in deposits of 1-5 m² at all locations during the sampling period except after the storm event on July 12 (Table 3.1.8.). E. coli concentrations in water and detached Cladophora are displayed in Figure 3.1.8. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ=0.061). E. coli levels in detached Cladophora were significantly higher than water concentrations (Mann-Whitney ρ <0.001). A statistically significant difference was not observed between beach locations for E. coli concentrations in detached Cladophora (Kruskal-Wallis $\rho = 0.0.046$). Station L was significantly different from C and R (Mann-Whitney ρ=0.0.002 and 0.038, respectively). Station L was the closest sampling location to a drainage ditch that enters South Linwood Beach. This station had the highest concentration of E. coli in detached Cladophora (4,888 cfu/g dwt).

Table 3.1.7. South Linwood Beach Water Quality Data 2008.

		E coli	in Water			Water	Chemistry	
Date		cfu/	100 ml		pН	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	рп	DO (IIIg/I)	remp C	μS/cm
06/23/08	1	< 1	1	1	7.92	7.7	21	420
07/01/08	1	2	15	6	8.16	7.7	22	433
07/08/08	1	2	2	1	8.15	7.6	23	439
07/15/08	2	1	1	1	8.16	7.6	23	448
07/22/08	1	1	10	4	8.19	7.6	24	439
07/29/08	35	66	76	59	8.11	7.6	24	446
08/05/08	1	1	1	1	8.11	7.6	24	431
08/12/08	1	2	3	2	8.14	7.7	24	431
Mean	5	11	14	9	8.12	7.6	23	436

Table 3.1.8. South Linwood Beach Cladophora Data 2008.

Date	E. c	oli in Cladop	ohora cfu/g	dwt	Cladophora	Microcystin	is in <i>Cladophora</i>	a μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	1412	390	716	839	4	<0.1	<0.1	<0.1
07/01/08	2452	60	190	901	4	<0.1	<0.1	<0.1
07/08/08	655	784	989	809	4	1.5	2.1	4
07/15/08	150	76	89	105	2	1.0	2.5	3
07/22/08	3324	2314	1964	2534	4	1.3	2.7	4
07/29/08	2353	756	812	1307	4	16.4	14.0	30
08/05/08	1325	533	834	897	4	17.0	24.0	41
08/12/08	4888	845	674	2136	4	35.0	43.0	78
Mean	2070	720	784	1191	4	12	15	27

(Cladophora Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all location; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } > 5 \text{ m}^2 \text{ in area}$.)

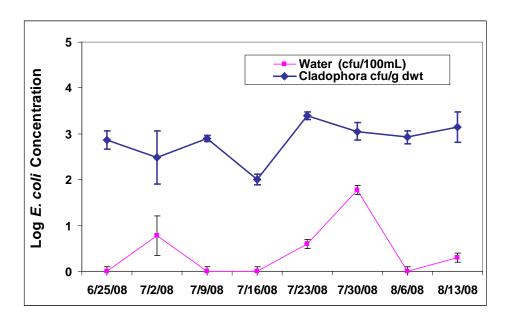


Figure 3.1.8. E. coli in Detached Cladophora and Water at South Linwood Beach 2008.

3.1.5. Pinconning Park

The sampling locations for Pinconning Park are provided in Figure 3.1.9. Three near-shore locations were sampled at depths of 0.3-0.5 m. The beach area was sandy and appeared to be groomed. Station L contained open water while Stations R and L contained *Typha* and *Scirpus*. Chara also was present along the lake bottom suggesting groundwater influx. Detached *Cladophora* was found only in isolated areas of the shallow littoral zone and the samples were collected approximately 1 meter from shore.

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.9. and 3.1.10., respectively. The mean *E. coli* concentration for the study period was 3 cfu/100 mL with no single beach sample exceeding 100 cfu/100 mL (Table 3.1.3.). Water temperatures ranged from 21°C to 24°C. The mean *E. coli* concentration in detached *Cladophora* was 299 cfu/g dwt (Table 3.1.4.). Microcystins were detected in the detached *Cladophora* beginning in July with mean concentrations of 0.7 μ g/g dwt, 3 μ g/g dwt, and 4 μ g/g dwt for microcystin RR, microcystin LR, and total microcystins, respectively. Detached *Cladophora* was present in isolated pockets at all locations for only 3 of the 8 sampling periods (Table 3.1.10.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.1.4. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.11). The drop in *E. coli* levels observed on July 15 was after a storm that had occurred on July 12. A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.073). Station C had the highest concentration of *E. coli* in detached *Cladophora* (1,454 cfu/g dwt).



Figure 3.1.9. Pinconning Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

Table 3.1.9. Pinconning Park Water Quality Data 2008.

		<i>E coli</i> ii	n Water			Water	Chemistry	
Date		cfu/1	00 ml		рН	Conductance		
	L	С	R	Mean	ριι	DO (mg/l)	Temp ^O C	μS/cm
06/23/08	1	2	1	1	7.56	7.7	21	460
07/01/08	1	2	1	1	8.16	7.6	22	464
07/08/08	2	1	3	2	8.17	7.7	23	463
07/15/08	2	1	3	2	8.10	7.6	23	475
07/22/08	1	1	1	1	8.15	7.7	24	474
07/29/08	4	8	9	7	8.12	7.6	24	466
08/05/08	4	2	< 1	2	8.11	7.6	24	470
08/12/08	1	2	9	4	8.15	7.7	24	470
Mean	2	2	3	3	8.06	7.7	23	468

Table 3.1.10. Pinconning Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E. (coli in Cladop	ohora cfu/g	dwt	Cladophora	Microcystins	in Cladophor	a µg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	NC	NC	NC	NC	0	NA	NA	NA
07/01/08	NC	NC	NC	NC	0	NA	NA	NA
07/08/08	NC	NC	NC	NC	0	NA	NA	NA
07/15/08	NC	NC	NC	NC	0	NA	NA	NA
07/22/08	NC	NC	NC	NC	0	NA	NA	NA
07/29/08	50	64	40	51	2	0.8	1.2	2
08/05/08	634	1454	188	759	2	0.9	1.8	3
08/12/08	184	18	56	86	2	0.7	9.8	10
Mean	289	512	95	299	1	0.7	3	4

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } > 5 \text{ m}^2 \text{ in area}$.)

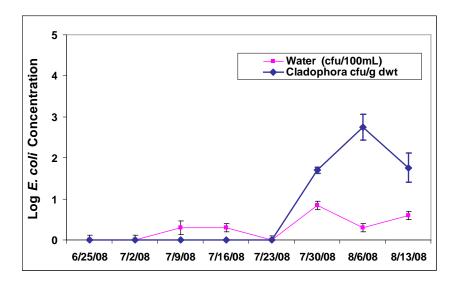


Figure 3.1.10. E. coli in Detached Cladophora and Water at Pinconning Park 2008.

3.1.6. White's Beach

The sampling locations for White's Beach are provided in Figure 3.1.11. Three near-shore locations were sampled at depths of 0.3-0.5 m. The beach area was sandy and appeared to be groomed. Station L contained open water while Stations R and C contained *Scirpus. Chara* also was present along the lake bottom suggesting groundwater influx. Detached *Cladophora* was only found in isolated areas of the shallow littoral zone and the samples were collected approximately 1 meter from shore.

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.11. and 3.1.12., respectively. The mean *E. coli* concentration for the study period was 16 cfu/100 mL with no single beach sample exceeding 100 cfu/100 mL (Table 3.1.11.). Water temperatures



Figure 3.1.11. White's Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)

ranged from 21°C to 24°C. Detached *Cladophora* was present in only one sample and the concentration of *E. coli* in the algae was 130 cfu/g dwt (Table 3.1.12.). Microcystin LR was detected in the detached *Cladophora* in only one sample in August (1.3 μ g/g dwt). Detached *Cladophora* was present only in the August 5 s ample at Station L (Table 3.1.10). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.1.12. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.007).

Table 3.1.11. White's Beach Water Quality Data 2008.

		E coli i	n Water			Water	Chemistry	
Date		cfu/1	00 ml		рН	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	ρп	DO (mg/i)	Temp ^o C	μS/cm
06/23/08	3	8	3	5	7.59	7.7	21	430
07/01/08	4	6	5	5	8.15	7.7	22	442
07/08/08	3	4	8	5	8.16	7.6	23	435
07/15/08	2	1	2	2	8.16	7.6	23	432
07/22/08	22	16	13	17	8.17	7.7	24	440
07/29/08	4	8	6	6	8.14	7.7	24	431
08/05/08	41	73	60	58	8.16	7.7	24	449
08/12/08	32	28	42	34	8.10	7.7	24	447
Mean	14	18	17	16	8.08	7.7	23	438

Table 3.1.12. White's Beach *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E.	coli in Cladophora	a cfu/g dwt		Cladophora	Microcystin	s in <i>Cladophora</i>	μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	NC	NC	NC	NC	0	NA	NA	NA
07/01/08	NC	NC	NC	NC	0	NA	NA	NA
07/08/08	NC	NC	NC	NC	0	NA	NA	NA
07/15/08	NC	NC	NC	NC	0	NA	NA	NA
07/22/08	NC	NC	NC	NC	0	NA	NA	NA
07/29/08	NC	NC	NC	NC	0	NA	NA	NA
08/05/08	130	NC	NC	_	1	NA	1.3	1.3
08/12/08	NC	NC	NC	NC	0	NA	NA	NA
Mean	130	NC	NC	_	0	NA		_

(Cladophora Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } 5 \text{ m}^2 \text{ in area}$.)

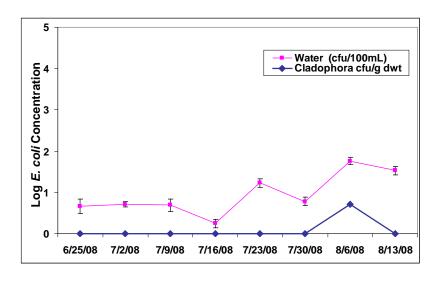


Figure 3.1.12. E. coli in Detached Cladophora and Water at White's Beach 2008.

3.1.7. Arenac County Park

The sampling locations for Arenac County Park are provided in Figure 3.1.13. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be groomed. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1 meter from shore.



Figure 3.1.13. Arenac County Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.13. and 3.1.14., respectively. The mean *E. coli* concentration for the study period was 25 cfu/100 mL with only one beach sample exceeding 100 c fu/100 mL (Station L, 111 cfu/100 mL; Table 3.1.3.). Water temperatures ranged from 21°C to 25°C. The mean *E. coli* concentration in

Table 3.1.13. Arenac County Park Water Quality Data 2008.

		E coli i	n Water			Wate	r Chemistry	,
Date		cfu/1	00 ml		Hq	Water Chemistry DO (mg/l) Temp °C Conductance μS/cm 11.5 19 310 10.0 19 300 10.6 20 284 10.0 20 296 8.0 21 315 11.2 21 280		Conductance
	L	С	R	Mean	рп	DO (IIIg/I)	19 19 20 20 21	μS/cm
06/23/08	5	2	2	3	8.13	11.5	19	310
07/01/08	45	31	94	57	8.17	10.0	19	300
07/08/08	55	42	29	42	8.20	10.6	20	284
07/15/08	2	1	9	4	8.15	10.0	20	296
07/22/08	1	3	<1	1	8.12	8.0	21	315
07/29/08	111	87	16	71	8.14	11.2	21	280
08/05/08	3	1	NC	1	8.11	11.4	21	292
08/12/08	1	2	60	21	8.17	11.3	21	296
Mean	28	21	26	25	8.15	11	20	297

Table 3.1.14. Arenac County Park Cladophora Data 2008.

Date	E.	coli in Cladopi	hora cfu/g c	lwt	Cladophora	Microcystii	ns in <i>Cladoph</i>	ora μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	1811	1144	2699	1885	4	<0.1	<0.1	<0.1
07/01/08	1801	1327	3258	2129	4	3	2	5
07/08/08	38	175	290	168	2	1	3	4
07/15/08	56	87	84	76	2	4	9	13
07/22/08	130	223	433	262	3	3	13	16
07/29/08	56	88	53	66	4	9	19	27
08/05/08	240	670	810	573	3	28	43	70
08/12/08	1300	1670	1890	1620	4	90	158	248
Mean	679	673	1190	847	3	20	35	55

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } 5 \text{ m}^2 \text{ in area}$.)

detached *Cladophora* was 847 cfu/g dwt (Table 3.1.14.). Microcystins were detected in the detached *Cladophora* beginning in July with mean concentrations of 20 μ g/g dwt, 35 μ g/g dwt, and 55 μ g/g dwt for microcystin RR, microcystin LR, and total microcystins, respectively. Detached *Cladophora* was present in 1-5 m p ockets at all locations for 6 of the 8 s ampling periods (Table 3.1.14.). The beach was groomed for the July 4th holiday and the *Cladophora* extent decreased to being present in isolated areas for two weeks. *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.1.14. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.61). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.57). S tation R had the highest concentration of *E. coli* in detached *Cladophora* (3,258 cfu/g dwt).

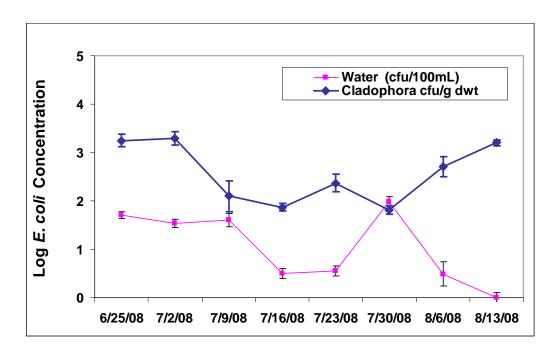


Figure 3.1.14. E. coli in Detached Cladophora and Water at Arenac County Park 2008.

3.1.8. Foster Road Beach

The sampling locations for Foster Road Beach are provided in Figure 3.1.15. Three near-shore locations were sampled at a depth of 0.5 m. The beach area was sandy and appeared to be groomed. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1 meter from shore. Strong wave action was observed at this location which kept the detached *Cladophora* suspended in the water column. This was the only Saginaw Bay location where suspended *Cladophora* was observed.

The results of water and detached *Cladophora* analyses are summarized in Tables 3.1.15. and 3.1.16., respectively. The mean *E. coli* concentration for the study period was 73 cfu/100 mL with only one set of beach samples averaging 315 c fu/100mL (July 15; Table 3.1.17.). This sample was taken after the storm event which occurred on July 12. Water temperatures ranged from 21°C to 25°C. The mean *E. coli* concentration in detached *Cladophora* was 2,169 cfu/g dwt (Table 3.1.16.). Microcystins were detected in the detached *Cladophora* beginning in July with mean concentrations of 36 μ g/g dwt, 41 μ g/g dwt, and 77 μ g/g dwt for microcystin RR, microcystin LR, and total microcystins, respectively. D etached *Cladophora* was present throughout the water column during the first three weeks, not present on July 15 (after the storm), and present only in isolated pockets during the remainder of the sampling period (Table 3.1.16.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.1.18. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.78). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.90). Station L had the highest concentration of *E. coli* in detached *Cladophora* (7,332 cfu/g dwt).

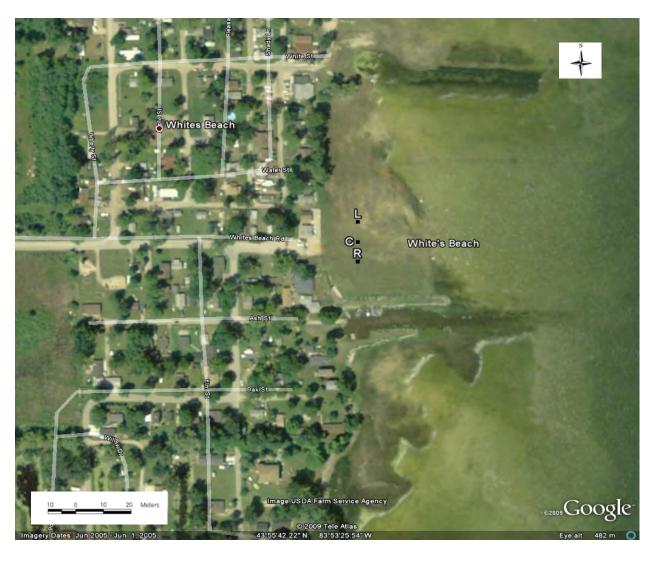


Figure 3.1.15. Foster Road Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)

Table 3.1.15. Foster Road Beach Water Quality Data 2008.

		E coli i	n Water		Water Chemistry				
Date		cfu/1	00 ml		рН	DO (mg/l)	Temp ^O C	Conductance	
	L	С	R	Mean	ρп	DO (mg/i)	Temp ^o C	μS/cm	
06/23/08	1	2	5	3	7.92	7.7	21	376	
07/01/08	60	21	23	35	8.18	7.6	22	382	
07/08/08	55	140	181	125	8.15	7.5	23	442	
07/15/08	320	400	225	315	8.15	7.4	23	436	
07/22/08	1	2	3	2	8.12	7.3	24	447	
07/29/08	25	31	50	35	8.20	7.7	24	436	
08/05/08	22	21	7	17	8.14	7.8	24	447	
08/12/08	46	51	71	56	8.11	8.1	24	450	
Mean	66	84	71	73	8.12	7.6	23	427	

Table 3.1.16. Foster Road Beach *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E. c	oli in Clado _l	ohora cfu/g	dwt	Cladophora	Microcystins	in <i>Cladophora</i>	μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	1380	2760	1515	1885	5	<0.1	<0.1	<0.1
07/01/08	4554	7010	5346	5637	5	<0.1	1	1
07/08/08	6731	5893	7332	6652	5	12	11	23
07/15/08	NC	NC	NC	NC	0	NA	NA	NA
07/22/08	150	230	320	233	2	27	34	61
07/29/08	230	156	240	209	2	68	80	148
08/05/08	213	270	189	224	2	80	80	161
08/12/08	332	380	320	344	2	82	102	184
Mean	1941	2386	2180	2169	3	36	41	77

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2$ in area; $4 = \text{Deposits } 1 - 5 \text{ m}^2$ in area; $5 = \text{Deposits } > 5 \text{ m}^2$ in area.)

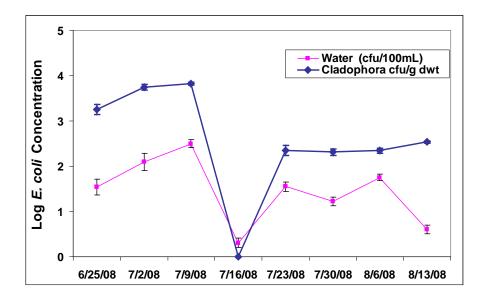


Figure 3.1.16. E. coli in Detached Cladophora and Water at Foster Road Beach 2008.

3.2 Grand Traverse Bay and Little Traverse Bay *E. coli*, Microcystin, and Water Chemistry Results

3.2.1. Petoskey State Park

The sampling locations for Petoskey State Park are provided in Figure 3.2.1. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be natural. Detached *Cladophora* was present in isolated pockets in the shallow littoral zone and the samples were collected approximately 1 meter from shore.



Figure 3.2.1. Petoskey State Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.1. and 3.2.2., respectively. The mean *E. coli* concentration for the study period was 13 cfu/100 mL with no single beach sample exceeding 100 cfu/100 mL (Table 3.2.1.). Water temperatures ranged from 19°C to 21°C. The mean *E. coli* concentration in detached *Cladophora* was 1,117 cfu/g dwt (Table 3.2.2.). Microcystins were not detected in the detached *Cladophora*. Detached *Cladophora* was present in isolated pockets at all locations for only 2 of the 8 sampling periods (Table 3.2.2.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.2.2. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.59). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.058). Station R had the highest concentration of *E. coli* in detached *Cladophora* (1,600 cfu/g dwt).

Table 3.2.1. Petoskey State Park Water Quality Data 2008.

		<i>E coli</i> ii	n Water			Wate	r Chemistry	
Date		cfu/1	00 ml		Hq	DO (mg/l)	Water Chemistry OO (mg/l) Temp OC μS/cm Conductance μS/cm 10.5 19 303 10.9 19 296 11.0 19 300 10.5 20 295 10.4 21 300 10.8 21 298	
	L	С	R	Mean	рп	DO (mg/i)	remp C	μS/cm
06/23/08	6	3	2	4	8.00	10.5	19	303
07/01/08	89	55	73	72	8.23	10.9	19	296
07/08/08	2	3	3	3	8.13	11.0	19	300
07/15/08	3	6	6	5	8.15	10.5	20	295
07/22/08	4	7	5	5	8.24	10.4	21	300
07/29/08	16	12	5	11	8.18	10.8	21	298
08/05/08	1	1	1	1	8.14	10.4	21	300
08/12/08	6	3	1	4	8.07	10.7	21	297
Mean	16	11	12	13	8.14	10.6	20	299

Table 3.2.2. Petoskey State Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E. coli	in Cladophora cfu	/g dwt		Cladophora	Microcystin	is in <i>Cladopho</i>	ra μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	990	1200	1100	1097	2	<0.1	<0.1	<0.1
07/01/08	1455	1390	1543	1463	2	<0.1	<0.1	<0.1
07/08/08	NC	NC	1200	1200	1	<0.1	<0.1	<0.1
07/15/08	NC	1423	NC	1423	1	<0.1	<0.1	<0.1
07/22/08	NC	556	NC	556	1	<0.1	<0.1	<0.1
07/29/08	NC	NC	1600	1600	1	<0.1	<0.1	<0.1
08/05/08	1200	NC	NC	1200	1	<0.1	<0.1	<0.1
08/12/08	NC	NC	400	400	1	<0.1	<0.1	<0.1
Mean	1215	1142	1169	1117	1	<0.1	<0.1	<0.1

(Cladophora Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } 5 \text{ m}^2 \text{ in area}$.)

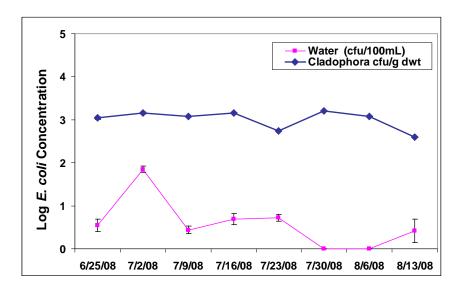


Figure 3.2.2. E. coli in Detached Cladophora and Water at Petoskey State Park 2008.

3.2.2. Magnus Park

The sampling locations for Magnus Park are provided in Figure 3.2.3. The beach area was sandy and appeared to be natural. Detached *Cladophora* was present in isolated pockets in the shallow littoral zone and the samples were collected approximately 1 meter from shore.



Figure 3.2.3. Magnus Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.3. and 3.2.4., respectively. The mean *E. coli* concentration for the study period was 17 cfu/100 mL with one beach sample exceeding 100 c fu/100 mL (120 cfu/100 mL; Table 3.2.3.). Water temperatures ranged from 19°C to 21°C. The mean *E. coli* concentration in detached *Cladophora* was 1,554 cfu/g dwt (Table 3.2.4.). Microcystins were not detected in the detached *Cladophora*. Detached *Cladophora* was present in isolated pockets at one location for 4 of the 8 sampling periods (Table 3.2.4.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.2.4. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.59). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.054). Station R had the highest concentration of *E. coli* in detached *Cladophora* (2,368 cfu/g dwt).

Table 3.2.3. Magnus Park Water Quality Data 2008.

		<i>E coli</i> ii	n Water			Wate	r Chemistry	,
Date		cfu/1	00 ml		На	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	рп	DO (mg/i)	Temp C	μS/cm
06/23/08	5	3	2	3	8.03	10.8	19	295
07/01/08	2	3	2	2	8.13	10.5	19	297
07/08/08	77	120	76	91	8.17	11.0	19	305
07/15/08	1	3	6	3	8.04	10.6	20	301
07/22/08	6	3	2	4	8.13	11.0	21	299
07/29/08	9	3	11	8	8.21	10.8	21	295
08/05/08	4	16	15	12	8.24	11.0	21	297
08/12/08	18	10	15	14	8.10	10.2	21	300
Mean	15	20	16	17	8.13	10.7	20	299

Table 3.2.4. Magnus Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E. coli in Cladophora cfu/g dwt				Cladophora	Microcystins in Cladophora μg/g dwt		
	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	2368	NC	3321	2845	1	<0.1	<0.1	<0.1
07/01/08	NC	NC	NC	NC	0	NC	NC	NC
07/08/08	NC	NC	1200	1200	1	<0.1	<0.1	<0.1
07/15/08	NC	NC	NC	NC	0	NC	NC	NC
07/22/08	NC	NC	NC	NC	0	NC	NC	NC
07/29/08	NC	NC	1300	1300	1	<0.1	<0.1	<0.1
08/05/08	NC	NC	NC	NC	0	NC	NC	NC
08/12/08	NC	NC	870	870	1	<0.1	<0.1	<0.1
Mean	2368	NC	1673	1554	1	<0.1	<0.1	<0.1

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } 5 \text{ m}^2 \text{ in area}$.)

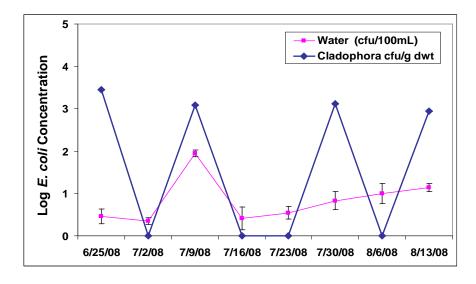


Figure 3.2.4. E. coli in Detached Cladophora and Water at Magnus Park 2008.

3.2.3. Elk Rapids Park

The sampling locations for Elk Rapids Park are provided in Figure 3.2.5. The beach area was sandy and appeared to be natural. Detached *Cladophora* was present in isolated pockets in the shallow littoral zone and the samples were collected approximately 1 meter from shore.



Figure 3.2.5. Elk Rapids Park Sampling Locations 2008. (L= Left, C=Center, R=Right).

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.5. and 3.2.6., respectively. The mean *E. coli* concentration for the study period was 74 cfu/100 mL with two sets of beach samples exceeding 100 c fu/100 mL (134 cfu/100 mL and 292 c fu/100 mL; Table 3.2.5.). Water temperatures ranged from 19°C to 21°C. The mean *E. coli* concentration in detached *Cladophora* was 1,225 cfu/g dwt (Table 3.2.6.). Microcystins were not detected in the detached *Cladophora*. Detached *Cladophora* was present in isolated pockets at more than one location for 6 of the 8 sampling periods (Table 3.1.6.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.1.4. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.18). A statistically significant difference was observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.030). Station R was significantly different from C and L (Mann-Whitney ρ =0.0.05 and 0.003, respectively). Station R was the closest sampling location

Table 3.2.5. Elk Rapids Park Water Quality Data 2008.

		E coli ii	n Water			Water	Chemistry	
Date		cfu/1	00 ml		pН	DO (mg/l)	Temp ^O C	Conductance
	┙	С	R	Mean	Pii	DO (mg/i)	Temp C	μS/cm
06/23/08	155	390	330	292	8.27	10.0	19	297
07/01/08	23	16	54	31	8.14	10.9	19	301
07/08/08	44	13	27	28	8.11	10.8	19	297
07/15/08	22	56	70	49	8.13	10.9	20	301
07/22/08	57	25	22	35	8.12	10.1	21	275
07/29/08	90	145	166	134	8.05	10.9	21	297
08/05/08	4	1	21	9	8.01	10.7	21	281
08/12/08	25	19	7	17	8.10	10.2	21	302
Mean	53	83	87	74	8.12	11	20	294

Table 3.2.6. Elk Rapids Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E. coli	in <i>Cladopho</i>	ora cfu/g dw	rt	Cladophora	Microcystin	s in <i>Cladopho</i>	ra μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	724	4111	6900	3912	2	<0.1	<0.1	<0.1
07/01/08	322	858	3754	1645	2	<0.1	<0.1	<0.1
07/08/08	NC	NC	770	770	1	<0.1	<0.1	<0.1
07/15/08	NC	NC	840	840	1	<0.1	<0.1	<0.1
07/22/08	569	670	440	560	2	<0.1	<0.1	<0.1
07/29/08	430	1100	1600	1043	2	<0.1	<0.1	<0.1
08/05/08	500	700	344	515	2	<0.1	<0.1	<0.1
08/12/08	330	448	780	519	2	<0.1	<0.1	<0.1
Mean	479	1314	1928	1225	2	<0.1	<0.1	<0.1

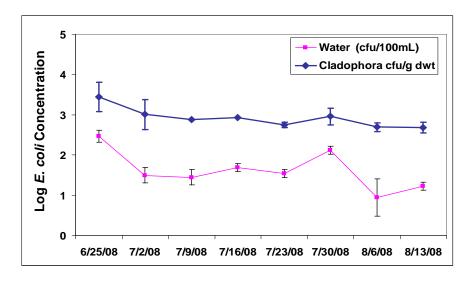


Figure 3.2.6. E. coli in Detached Cladophora and Water at the Elk Rapids Park 2008.

location to the creek which drains a duck pond ne ar the city. Station R had the highest concentration of *E. coli* in detached *Cladophora* (6,900 cfu/g dwt).

3.2.4. Antrim County Day Park

The sampling locations for Antrim County Day Park are provided in Figure 3.2.7. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be natural. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1 meter from shore.



Figure 3.2.7. Antrim County Day Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.7. and 3.2.8., respectively. The mean *E. coli* concentration for the study period was 25 cfu/100 mL with one beach sample exceeding 100 c fu/100 mL (111 cfu/100 mL; Table 3.2.7.). W ater

temperatures ranged from 19°C to 21°C. The mean $E.\ coli$ concentration in detached Cladophora was 350 cfu/g dwt (Table 3.2.8.). Microcystins were not detected in the detached Cladophora. Detached Cladophora was either absent or present in isolated pockets at one location for 7 of the 8 sampling periods and at 2 locations during 1 sampling period (Table 3.1.8.). $E.\ coli$ concentrations in water and detached Cladophora are displayed in Figure 3.2.8. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.68). A statistically significant difference was not observed between beach locations for $E.\ coli$ concentrations in detached Cladophora (Kruskal-Wallis ρ =0.27). S tation R had the highest concentration of $E.\ coli$ in detached Cladophora (660 cfu/g dwt).

Table 3.2.7. Antrim County Day Park Water Quality Data 2008.

		E coli i	n Water			Wate	r Chemistry	1
Date		cfu/1	00 ml		рН	DO (mg/l)	Town OC	Conductance
	L	С	R	Mean	ρп	DO (mg/i)	Temp °C	μS/cm
06/23/08	5	2	2	3	8.13	11.5	19	310
07/01/08	45	31	94	57	8.17	10.0	19	300
07/08/08	55	42	29	42	8.20	10.6	20	284
07/15/08	2	1	9	4	8.15	10.0	20	296
07/22/08	1	3	<1	1	8.12	8.0	21	315
07/29/08	111	87	16	71	8.14	11.2	21	280
08/05/08	3	1	NC	1	8.11	11.4	21	292
08/12/08	1	2	60	21	8.17	11.3	21	296
Mean	28	21	26	25	8.15	11	20	297

Table 3.2.8. Antrim County Day Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E.	coli in Cladophora	a cfu/g dwt		Cladophora	Microcys	tins in <i>Clad</i> d	ophora µg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	79	61	660	267	2	<0.1	<0.1	<0.1
07/01/08	NC	NC	NC	NC	0	NA	NA	NA
07/08/08	NC	NC	NC	NC	0	NA	NA	NA
07/15/08	NC	NC	NC	NC	0	NA	NA	NA
07/22/08	NC	NC	440	440	1	NA	<0.1	NA
07/29/08	NC	NC	NC	NC	0	NA	NA	NA
08/05/08	NC	NC	344	344	1	NA	<0.1	NA
08/12/08	NC	NC	NC	NC	0	NA	NA	NA
Mean	_	_	481	350	1	_	<0.1	_

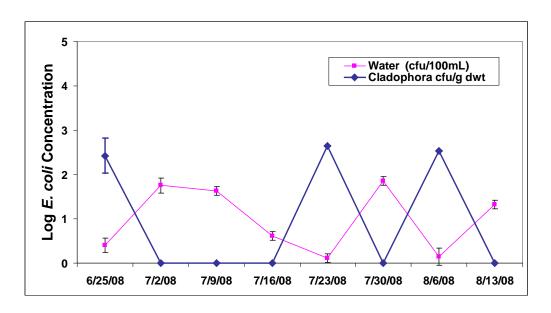


Figure 3.2.8. *E. coli* in Detached *Cladophora* and Water at the Antrim County Day Park 2008.

3.2.5. East Bay Park

The sampling locations for East Bay Park are provided in Figure 3.2.9. Three near-shore locations were sampled at depths of 0.3-0.5 m. The beach area was sandy and appeared to be natural. Station L contained open water while Stations R and C contained *Scirpus*. Detached *Cladophora* was found at all of the shallow littoral zone locations and the samples were collected approximately 1 meter from shore.

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.9. and 3.2.10., respectively. The mean *E. coli* concentration for the study period was 25 cfu/100 mL with one sample exceeding 100 cfu/100 mL (103 cfu/100 mL; Table 3.2.9.). Water temperatures ranged from 19°C to 21°C. The mean *E. coli* concentration in detached *Cladophora* was 969 cfu/g dwt (Table 3.2.10.). M icrocystins were not detected in the detached *Cladophora*. Detached *Cladophora* was present in isolated pockets at more than one location for all of the 8 sampling periods (Table 3.2.10.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.2.10. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.12). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.41). Station L had the highest *E. coli* concentration in detached *Cladophora* (5,000 cfu/g dwt). Station L was the closest sampling location to a storm-water outlet entering Grand Traverse Bay.



Figure 3.2.9. East Bay Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

Table 3.2.9. East Bay Park Water Quality Data 2008.

		E coli ii	n Water			Water	Chemistry	
Date		cfu/1	00 ml		Hq	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	рп	DO (IIIg/I)	Temp C	μS/cm
06/23/08	2	1	3	2	8.21	10.6	19	299
07/01/08	5	3	NC	3	8.21	10.4	19	300
07/08/08	4	7	14	8	8.07	10.5	19	320
07/15/08	17	41	21	26	8.07	10.3	20	310
07/22/08	41	91	103	78	8.04	10.1	21	330
07/29/08	51	92	61	68	8.19	10.2	21	324
08/05/08	8	4	11	8	8.05	10.3	21	309
08/12/08	7	2	1	8	8.09	10.8	21	317
Mean	17	30	27	25	8.12	10	20	314

Table 3.2.10. East Bay Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	Е. с	coli in Cladop	hora cfu/g dv	wt	Cladophora	Microcystins	in Cladophora	μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	5000	2000	1000	2667	3	<0/1	<0/1	<0/1
07/01/08	3300	1500	900	1900	3	<0/1	<0/1	<0/1
07/08/08	660	500	460	540	3	<0/1	<0/1	<0/1
07/15/08	546	600	NC	573	2	<0/1	<0/1	<0/1
07/22/08	340	500	600	480	3	<0/1	<0/1	<0/1
07/29/08	700	630	NC	665	2	<0/1	<0/1	<0/1
08/05/08	935	500	700	712	3	<0/1	<0/1	<0/1
08/12/08	325	330	NC	218	2	<0/1	<0/1	<0/1
Mean	1476	820	732	969	3	<0/1	<0/1	<0/1

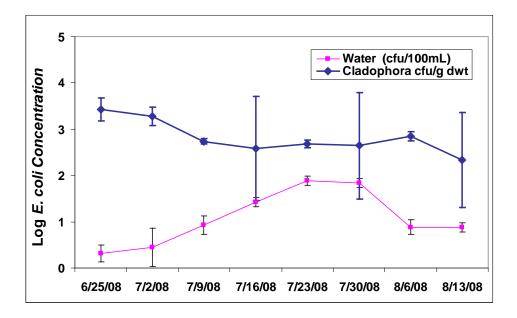


Figure 3.2.10. E. coli in Detached Cladophora and Water at East Bay Park 2008.

3.2.6. Traverse City State Park

The sampling locations for Traverse City State Park are provided in Figure 3.2.11. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be natural. Detached *Cladophora* was present in isolated pockets in the shallow littoral zone near Mitchell Creek and the samples were collected approximately 1 meter from shore.



Figure 3.2.11. Traverse City State Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.11. and 3.2.12., respectively. The mean *E. coli* concentration for the study period was 33 cfu/100 mL with one sample exceeding 100 cfu/100 mL (195 cfu/100 mL; Table 3.2.11.). W ater temperatures ranged from 19°C to 21°C. Station L was the only location at this beach with detached Cladophora and the associated *E. coli* concentrations averaged 1,630 cfu/g dwt. The highest concentration *E. coli* in detached *Cladophora* was 3,300 cfu/g dwt. This station was the closest sampling location to Mitchell Creek which has a history of bacterial contamination problems (The Watershed Center 2006). Microcystins were not detected in the detached *Cladophora*. Detached *Cladophora* was present in isolated pockets at one location for all of the 8 sampling periods (Table 3.2.12.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.2.12. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.15). A statistically significant difference was observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.001).

Table 3.2.11. Traverse City State Park Water Quality Data 2008.

		E coli i	n Water			Water	Chemistry	
Date		cfu/1	00 ml		рН	DO (mg/l)	Tomp OC	Conductance
	L	С	R	Mean	рп	DO (mg/i)	Temp C	μS/cm
06/23/08	1	4	NC	2	8.16	10.7	19	301
07/01/08	3	5	10	6	8.15	10.1	19	305
07/08/08	195	37	66	99	8.15	10.5	19	296
07/15/08	22	48	63	44	8.20	10.4	20	297
07/22/08	95	88	57	80	8.01	10.8	21	299
07/29/08	19	10	5	11	8.02	10.2	21	300
08/05/08	18	14	11	14	8.25	10.8	21	294
08/12/08	8	6	5	6	8.05	10.6	21	294
Mean	45	27	31	33	8.12	10.5	20	298

Table 3.2.12. Traverse City State Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E.	coli in Cladophora	a cfu/g dwt		Cladophora	Microcystins	in Cladoph	ora μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	2000	NC	NC	_	1	<0/1	<0/1	<0/1
07/01/08	3300	NC	NC	_	1	<0/1	<0/1	<0/1
07/08/08	900	NC	NC	_	1	<0/1	<0/1	<0/1
07/15/08	750	NC	NC	_	1	<0/1	<0/1	<0/1
07/22/08	1200	NC	NC	_	1	<0/1	<0/1	<0/1
07/29/08	700	NC	NC	_	1	<0/1	<0/1	<0/1
08/05/08	990	NC	NC	_	1	<0/1	<0/1	<0/1
08/12/08	3200	NC	NC	_	1	<0/1	<0/1	<0/1
Mean	1630	NC	NC		1	<0/1	<0/1	<0/1

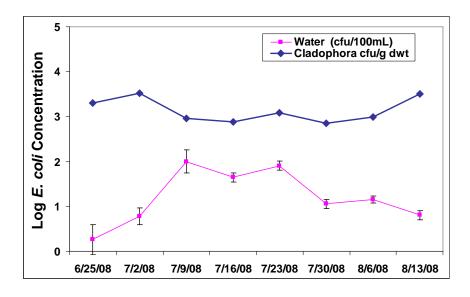


Figure 3.2.12. E. coli in Detached Cladophora and Water at Traverse City State Park 2008.

3.2.7. Bryant Park

The sampling locations for Bryant Park are provided in Figure 3.2.13. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be natural. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1 meter from shore.



Figure 3.2.13. Bryant Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.13. and 3.2.14., respectively. The mean *E. coli* concentration for the study period was 46 cfu/100 mL with one set of samples exceeding 100 cfu/100 mL (281 cfu/100 mL; Table 3.2.13.). Water temperatures ranged from 19°C to 21°C. The mean *E. coli* concentration in detached *Cladophora* was 1,873 cfu/g dwt (Table 3.2.14.). Microcystins were not detected in the detached *Cladophora*. Detached *Cladophora* was present in isolated pockets at one location for all of the 8 sampling periods (Table 3.2.14.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.2.14. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.82). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.48). Station C had the highest concentration of *E. coli* in detached *Cladophora* (9,460 cfu/g dwt).

Table 3.2.13. Bryant Park Water Quality Data 2008.

		E coli ii	n Water			Wate	r Chemistry	
Date		cfu/1	00 ml		рН	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	рп	DO (IIIg/I)	remp C	μS/cm
06/23/08	3	4	11	6	8.25	10.5	19	296
07/01/08	6	8	5	6	8.00	10.2	19	301
07/08/08	290	310	243	281	8.24	10.0	19	300
07/15/08	8	9	4	7	8.19	10.6	20	298
07/22/08	6	8	3	6	8.13	10.3	21	300
07/29/08	11	13	22	15	8.11	10.4	21	297
08/05/08	12	18	15	15	8.15	10.7	21	317
08/12/08	33	29	1	28	8.05	10.1	21	299
Mean	46	50	38	46	8.14	10.3	20	301

Table 3.2.14. Bryant Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E	E. coli in Cladophor	a cfu/g dwt		Cladophora	Microcystins	s in <i>Cladoph</i> o	ora µg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	980	9460	680	3707	2	<0.1	<0.1	<0.1
07/01/08	977	7690	490	3052	2	<0.1	<0.1	<0.1
07/08/08	3443	1462	2741	2549	2	<0.1	<0.1	<0.1
07/15/08	1300	1800	1567	1556	2	<0.1	<0.1	<0.1
07/22/08	990	832	751	858	2	<0.1	<0.1	<0.1
07/29/08	111	1643	1565	1106	2	<0.1	<0.1	<0.1
08/05/08	1245	890	1443	1193	2	<0.1	<0.1	<0.1
08/12/08	1122	844	922	963	2	<0.1	<0.1	<0.1
Mean	1271	3078	1270	1873	2	<0.1	<0.1	<0.1

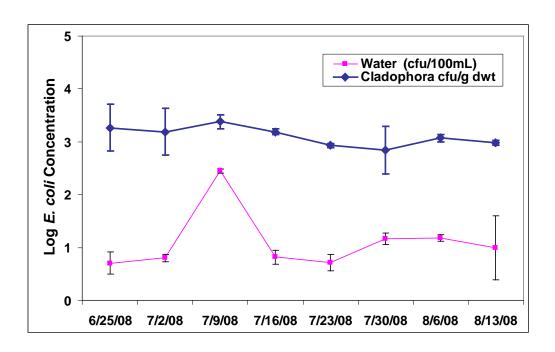


Figure 3.2.14. E. coli in Detached Cladophora and Water at Bryant Park 2008.

3.2.8. Clinch Park

The sampling locations for Clinch Park are provided in Figure 3.2.15. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be natural. Detached *Cladophora* was present at one location during the last sampling event. The sample was collected approximately 0.5 m from shore.

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.15. and 3.2.16., respectively. The mean *E. coli* concentration for the study period was 35 cfu/100 mL with one set of samples exceeding 100 cfu/100 mL (164 cfu/100 mL; Table 3.2.15.). Water temperatures ranged from 19°C to 21°C. Detached *Cladophora* was present in an isolated pocket at one location during the August 12 sampling event (Table 3.2.16.). *E. coli* was present in detached *Cladophora* in this sample at a concentration of 680 c fu/g dwt (Table 3.2.16.). Microcystins were not detected in the detached *Cladophora*. *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.2.16. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.99).



Figure 3.2.15. Clinch Park Sampling Locations 2008. (L= Left, C=Center, R=Right)

Table 3.2.15. Clinch Park Water Quality Data 2008.

		E coli ii	n Water			Water	Chemistry	
Date		cfu/1	00 ml		рН	DO (mg/l)	Temp ^O C	Conductance
	L	С	R	Mean	рп	DO (IIIg/I)	Temp C	μS/cm
06/23/08	22	14	11	16	8.28	10.5	19	296
07/01/08	1	3	NC	1	8.01	10.9	19	300
07/08/08	33	19	30	27	8.05	10.1	19	310
07/15/08	21	18	32	24	8.16	10.7	20	302
07/22/08	155	170	167	164	8.02	10.3	21	302
07/29/08	43	21	37	34	8.01	10.4	21	307
08/05/08	11	7	18	12	8.30	10.2	21	299
08/12/08	4	2	1	3	8.22	10.4	21	298
Mean	36	32	37	35	8.13	10.4	20	302

Table 3.2.16. Clinch Park *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date	E.	coli in Cladophora	a cfu/g dwt		Cladophora	Microcystins	s in <i>Cladopho</i>	ra μg/g dwt
Date	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	NC	NC	NC	NC	0	<0.1	<0.1	<0.1
07/01/08	NC	NC	NC	NC	0	<0.1	<0.1	<0.1
07/08/08	NC	NC	NC	NC	0	<0.1	<0.1	<0.1
07/15/08	NC	NC	NC	NC	0	<0.1	<0.1	<0.1
07/22/08	NC	NC	NC	NC	0	<0.1	<0.1	<0.1
07/29/08	NC	NC	NC	NC	0	<0.1	<0.1	<0.1
08/05/08	NC	NC	NC	NC	0	<0.1	<0.1	<0.1
08/12/08	NC	NC	680	680	1	<0.1	<0.1	<0.1
Mean	NC	NC	680	680	0	<0.1	<0.1	<0.1

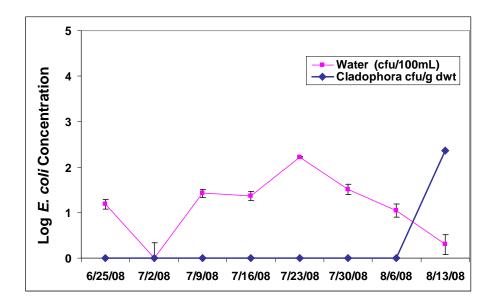


Figure 3.2.16. E. coli in Detached Cladophora and Water at Clinch Park 2008.

3.2.9. West End Beach

The sampling locations for West End Beach are provided in Figure 3.2.17. Three near-shore locations were sampled at a depth of 0.3 m. The beach area was sandy and appeared to be natural. Detached *Cladophora* was common in the shallow littoral zone and the samples were collected approximately 1 meter from shore. This beach appeared to be used by pet owners, as dogs were frequently present during sampling.

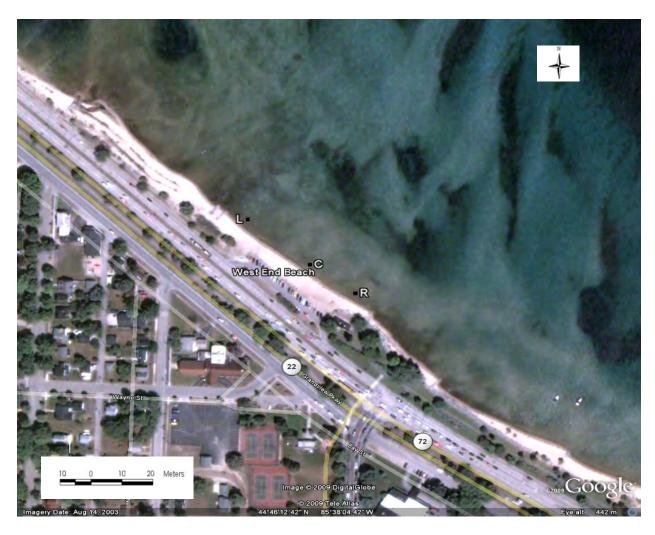


Figure 3.2.17. West End Beach Sampling Locations 2008. (L= Left, C=Center, R=Right)

The results of water and detached *Cladophora* analyses are summarized in Tables 3.2.17. and 3.2.18., respectively. The mean *E. coli* concentration for the study period was 35 cfu/100 mL with no samples exceeding 100 c fu/100 mL (Table 3.2.17.). Water temperatures ranged from 19°C to 21°C. The mean *E. coli* concentration in detached *Cladophora* was 2,976 cfu/g dwt (Table 3.2.18.). Microcystins were not detected in the detached *Cladophora*. Detached *Cladophora* was present in isolated pockets at all locations for 4 of the 8 sampling periods (Table 3.2.18.). *E. coli* concentrations in water and detached *Cladophora* are displayed in Figure 3.2.18. No correlation was observed between bacterial levels in water and detached algae (Spearman's RO ρ =0.28). A statistically significant difference was not observed between beach locations for *E. coli* concentrations in detached *Cladophora* (Kruskal-Wallis ρ =0.79). Station C was higher than the other stations with respect to mean *E. coli* concentrations in detached *Cladophora* (3078 cfu/g dwt). The highest concentration of *E. coli* in detached *Cladophora* measured during the study period in Grand Traverse Bay was at West End Beach (22,874 cfu/g dwt at Station C). Dogs frequently were present on this beach at the time of sampling, which may be the source of the high bacteria concentrations.

Table 3.2.17. West End Beach Water Quality Data 2008.

	<i>E coli</i> in Water				Water Chemistry				
Date	cfu/100 ml				На	DO (mg/l)	Temp ^O C	Conductance	
	L	С	R	Mean	ριι	DO (IIIg/I)	remp C	μS/cm	
06/23/08	5	3	4	4	8.21	10.3	19	302	
07/01/08	3	1	2	2	8.29	11.0	19	297	
07/08/08	34	55	48	46	8.07	10.0	19	303	
07/15/08	46	33	25	35	8.03	10.8	20	301	
07/22/08	66	86	126	93	8.04	10.5	21	297	
07/29/08	6	14	5	8	8.25	10.6	21	294	
08/05/08	100	94	49	81	8.27	10.7	21	306	
08/12/08	22	10	1	14	8.27	10.7	21	297	
Mean	35	37	33	35	8.18	10.6	20	300	

Table 3.2.18. West End Beach *Cladophora* Data 2008. (NC=No *Cladophora* Present; NA=Not Analyzed)

Date -	E. coli in Cladophora cfu/g dwt				Cladophora	Microcystins in Cladophora μg/g dwt		
	L	С	R	Mean	Extent	RR	LR	Total
06/23/08	1509	76	289	625	2	<0.1	<0.1	<0.1
07/01/08	1721	22874	13337	12644	2	<0.1	<0.1	<0.1
07/08/08	1016	NC	NC	1016	1	<0.1	<0.1	<0.1
07/15/08	NC	NC	1644	1644	1	<0.1	<0.1	<0.1
07/22/08	NC	NC	NC	NC	0	NA	NA	NA
07/29/08	NC	NC	NC	NC	0	NA	NA	NA
08/05/08	1300	1233	1567	1367	2	<0.1	<0.1	<0.1
08/12/08	678	444	567	563	2	<0.1	<0.1	<0.1
Mean	1245	6157	3481	2976	1	<0.1	<0.1	<0.1

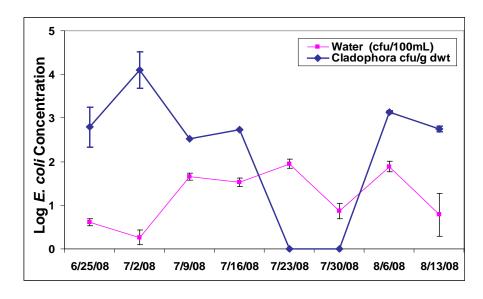


Figure 3.2.18. E. coli in Detached Cladophora and Water at West End Beach 2008.

3.3 Saginaw Bay and Grand Traverse Bay/Little Traverse Bay Comparisons

A summary of the project data is presented in Table 3.3.1. Mean temperatures for Saginaw Bay were higher than in Grand Traverse Bay/Little Traverse Bay (23°C and 20°C, respectively). Microcystins in detached *Cladophora* were absent in Grand Traverse Bay/Little Traverse Bay. In Saginaw Bay, the grand mean of all locations with detached *Cladophora* was 57 μ g/g dwt for the study period. Mean concentrations of *E. coli* in water were significantly higher (Mann-Whitney ρ =0.003) in Grand Traverse Bay/Little Traverse Bay (34 cfu/100 mL) than in Saginaw Bay (18 cfu/100 mL). In contrast, mean *E. coli* concentrations in detached *Cladophora* were higher in Saginaw Bay (2,796 cfu/g dwt) than Grand Traverse Bay/Little Traverse Bay (1,775 cfu/g dwt); however, the difference was not statistically significant (Mann-Whitney ρ =0.40). The mean *Cladophora* extent score for Grand Traverse Bay/Little Traverse Bay was 1, indicating that detached algae were found in small pockets. The mean *Cladophora* extent score for Saginaw Bay was 3 indicating that detached algae were found in 1 m² deposits.

Table 3.3.1. Summary of Water Quality and *Cladophora* Data for Grand Traverse Bay/Little Traverse Bay and Saginaw Bay 2008.

Parameter	Saginaw Bay	Grand Traverse/Little Traverse Bay		
Mean Temperature	23°C	20°C		
Mean Total Microcystins	57 μg/g dwt	<0.1 µg/g dwt		
Cladophora Extent Score	3	1		
Mean <i>E coli</i> in Water	18 cfu/100 mL	34 cfu/100 mL		
Mean E coli in Cladophora	2,796 cfu/g dwt	1,775 cfu/g dwt		

(*Cladophora* Extent: 0 = None Present; 1 = Present in isolated pockets at one location; 2 = Present in isolated pockets at all locations; $3 = \text{Deposits } 1 \text{ m}^2 \text{ in area}$; $4 = \text{Deposits } 1 - 5 \text{ m}^2 \text{ in area}$; $5 = \text{Deposits } 5 \text{ m}^2 \text{ in area}$.)

4.0 Discussion

The presence of Cladophora has been associated with high levels of E. coli and species of potentially pathogenic enteric bacteria in beach sand and swimming waters of the Great Lakes (Whitman et al. 2003; Ishii et al. 2006; Olapade et al. 2006). Enteric bacteria can grow within detached *Cladophora* under certain conditions (Byappanahalli et al. 2005). In addition, detached Cladophora deposits can potentially harbor and enhance the survival of pathogenic bacteria released into the environment through point and non-point sources (Byappanahalli et al. 2005). Recently, Clostridium was found to grow in detached Cladophora in the Great Lakes (Byappanahalli and Whitman 2009). Algal species, including *Cladophora*, have been reported to provide nutrients and to protect attached bacteria from environmental stresses, desiccation, predation, and harmful UV radiation (Byappanahalli et al. 2005). In this manner, Cladophora mats may play a significant role as a source and sink for pathogens in the near-shore environment. Based on the results from this investigation, Saginaw Bay appears to be more heavily impacted by detached Cladophora than Grand Traverse Bay/Little Traverse Bay. Eutrophication of lakes has been associated with increases in *Cladophora* mat production and accumulation (Stevenson et al. 2006; Herbst, 1969), but water transparency also has been suggested as an important causative factor (Barbiero et al. 2006; Bootsma, 2007). Elevated levels of nutrients in Saginaw Bay have been reported to stimulate blooms of cyanobacteria in Saginaw Bay and also influence the near-shore environment (Fahnenstiel et al. 2008; Millie et al.

2008). Differences in *Cladophora* accumulation between the Lake Huron and Lake Michigan sites may be attributed to higher total phosphorus levels (18 μ g/l vs. 5 μ g/l; MDNRE 2006). Levels of *E. coli* in detached *Cladophora* in both systems were similar to concentrations previously reported in the Great Lakes (3 \log_{10} - 5 \log_{10} ; Engelbert et al. 2008).

In Saginaw Bay, the highest levels of E. coli in detached Cladophora were consistently found at locations near the Saginaw River (Wenona Beach and the Bay City Recreation Area). Even within individual sites, locations near tributaries and drains at Wenona Beach and South Linwood Beach were significantly higher than locations farther away form a point source. This relationship also was noted in Grand Traverse Bay where the location near Mitchell Creek at the Traverse City State Park had elevated E. coli concentrations in detached Cladophora compared to the other two beach stations. At Traverse City State Park, Cladophora deposits were present only at the location near Mitchell Creek. These results suggest that *Cladophora* can trap bacteria from point sources and also be stimulated by nutrient discharges. Two locations, Pinconning Park and White's Beach had very limited Cladophora growth. Both locations had Chara growing on the lake bottom. Chara is known to exhibit allelopathic activity that can limit the growth of other aquatic plants (Berger and Schagerl 2004). No correlation was found between E. coli levels in the open water (designated beach monitoring locations) and the near-shore zone where the detached Cladophora samples were taken. As noted in previous studies, Cladophora appears to hold trapped E. coli and not release the entrained bacteria into the offshore water (Byappanahalli et al. 2003; Engelbert et al. 2008).

This investigation was the first to document the accumulation of microcystins in the detached *Cladophora* of Saginaw Bay. Saginaw Bay has a history of *Microcystis* blooms in the late summer months that produce both microcystin LR and RR (Fahnenstiel et al. 2008; Millie et al. 2008). Since *Microcystis* has a high requirement for sunlight, cyanobacteria may become stressed when they become trapped in the detached algae deposits. Stress has been implicated in initiating cyanotoxin production (Codd 1995). While accidental ingestion by humans of microcystins trapped in *Cladophora* is unlikely, these compounds can act as skin irritants (Bell and Codd 1994). Walking through *Cladophora* accumulations to get to deeper water may provide sufficient exposure to cause irritation in sensitive individuals if microcystins are present.

While all of the data suggest that swimming areas (1 m depth) are not impacted by *E. coli* accumulations in detached *Cladophora*, entrained bacteria and cyanotoxins may pose a hazard to children playing in the near-shore water and beach sand. While current regulations discourage beach grooming, the presence of elevated bacteria and microcystin levels in the near-shore environment of Saginaw Bay suggests that the current policy should balance potential impacts to public health with the ecosystem services provided by coastal wetlands.

5.0 Conclusions

An assessment of the ability of *Cladophora* mats to sequester *E. coli* and microcystin LR and RR was conducted in near-shore waters of Grand Traverse Bay (7 sites), Little Traverse Bay (2 sites), and Saginaw Bay (8 sites). The sampling locations were at public beach access points where *Cladophora* mats previously have been observed. The goals of this research were to determine the spatial and temporal variability of *E. coli* populations in *Cladophora* mats in these

recreational waters and if cyanotoxins (microcystin LR and RR) were sequestered in the detached algae. The collection of *Cladophora* samples was coordinated with local beach monitoring programs to facilitate the comparison with ambient water bacteria concentrations. This project provided important data for the assessment of public health impacts and the development of beach management programs to address the problems associated with *Cladophora* accumulations.

Based on the results from this investigation, Saginaw Bay appears to be more heavily impacted by detached Cladophora than Grand Traverse Bay/Little Traverse Bay. Mean E. coli concentrations in detached Cladophora were higher in Saginaw Bay (2,796 cfu/g dwt) than Grand Traverse Bay/Little Traverse Bay (1,775cfu/g dwt); however, the difference was not statistically significant (Mann-Whitney ρ =0.40). The mean *Cladophora* extent score for Grand Traverse Bay/Little Traverse Bay was 1, indicating that detached alga was consistently found in small pockets at a single location. The mean Cladophora extent score for Saginaw Bay was 3, indicating that detached alga was found in 1 m² deposits at all locations. Differences in Cladophora accumulation between Saginaw Bay and Grand Traverse Bay/Little Traverse Bay may be attributed to higher total phosphorus levels in Saginaw Bay. Levels of E. coli in detached Cladophora in both systems were similar to concentrations previously reported in the Great Lakes (1,000 cfu/g dwt – 60,000 cfu/g dwt). In Saginaw Bay, the highest levels of E. coli in detached Cladophora were consistently found at locations near the Saginaw River (Wenona Beach and the Bay City Recreation Area). Even within individual sites, locations near tributaries and drains at Wenona Beach and South Linwood Beach were significantly higher than locations farther away form a point source. This relationship also was noted in Grand Traverse Bay where the location near Mitchell Creek at the Traverse City State Park had elevated E. coli concentrations in detached *Cladophora* compared to the other two beach stations, where no detached algae was found. These results suggest that Cladophora can trap bacteria from point sources and also be stimulated by nutrient discharges. Two locations, Pinconning Park and White's Beach, had very limited Cladophora growth. Both locations had Chara growing on the lake bottom. Chara is known to exhibit allelopathic activity that can limit the growth of other aquatic plants. No correlation was found between E. coli levels in the open water (designated beach monitoring locations) and the near-shore zone where the detached *Cladophora* samples were taken. As noted in previous studies, Cladophora appears to hold trapped E. coli and does not release the entrained bacteria into the offshore water.

This investigation was the first to document the accumulation of microcystins in the detached *Cladophora* of Saginaw Bay. Total microcystins in detached *Cladophora* had a grand mean of 57 µg/g dwt for the study period. Saginaw Bay has a history of *Microcystis* blooms in the late summer months that produce both microcystin LR and RR. Since *Microcystis* has a high requirement for sunlight, cyanobacteria may become stressed when they become trapped in the detached algae mats. While accidental ingestion by humans of microcystins trapped in *Cladophora* is unlikely, these compounds can act as skin irritants. Walking through *Cladophora* accumulations to get to deeper water may provide sufficient exposure to cause irritation in sensitive individuals if microcystins are present. While all of the data suggest that swimming areas (1 m depth) are not impacted by *E. coli* accumulations in detached *Cladophora*, entrained bacteria and cyanotoxins may pose a hazard to children playing in the near-shore water and beach sand. Current regulations discourage beach grooming and altering the near-shore zone. The presence of elevated bacteria and microcystin levels in the near-shore environment of

Saginaw Bay suggest that the current policy should be reevaluated to balance potential impacts to public health with the ecosystem services provided by coastal wetlands.

6.0 References

- APHA (American Public Health Association). 1998. Standard Methods for the Evaluation of Water and Wastewater. 20th edition. APHA, Washington, DC.
- Barbiero, R.P., M.C. Tuchman, and E.S. Millard. 2006. Post-dreissenid increases in transparency during summer stratification in the offshore waters of Lake Ontario: Is a reduction in whiting events the cause? J. Great Lakes Res. 32:131–141.
- Bartsch, A. F. 1968. Eutrophication is beginning in Lake Michigan. 1968. Water Waste Eng. 5: 84-87.
- Bell, S.G and Codd, G. A. 1994. Cyanobacterial toxins and human health. Rev Med Microbiol. 5:256–264.
- Berger, J. and Schagerl, M. 2004. Allelopathic activity of Characeae. Biologia 59:9–15.
- Bootsma, H.A., E.B. Young, and J.A. Berges (eds.). 2005. *Cladophora* research and management in the Great Lakes. Proceedings of workshop, December 2004. Special Report No. 2005-01, UWM Great Lakes WATER Institute.
- Boyer, G., M. C. Watzin, A. D. Shambaugh, M. F. Satchwell, B. R. Rosen, and T. Mihuc. 2004. The occurrence of cyanobacterial toxins in Lake Champlain. In: "Lake Champlain: partnerships and Research in the New Millennium. (Proceedings of the Lake Champlain Research Consortium, May 20th 2002, Saint-Jean-sur-Richelieu, Quebec" T. Manley, Ed., p 241-257.
- Byappanahalli, M. N., D. A. Shively, M. B. Nevers, M. J. Sadowsky, and R. L. Whitman. 2003. Growth and survival of *Escherichia coli* and enterococci populations in the macro-alga *Cladophora* (Chlorophyta). FEMS Microbiol. Ecol. 46:203-211.
- Byappanahalli, M. N. and R. L. Whitman. 2009. Clostridium botulinum type E occurs and grows in the alga *Cladophora* glomerata. Can. J. Fish. Aquat. Sci. 66: 879–882
- Codd, G.A. 1995. Cyanobacterial toxins: occurrence, properties and biological significance. Water Sci. Technol. 32, 149^156.
- Dodds, W. K., and D. A. Gudder. 1992. The ecology of *Cladophora*. J. Phycol. 28:415-427
- Englebert E.T., McDermott C., and G.T. Kleinheinz. 2008. Effects of the nuisance algae, *Cladophora*, on Escherichia coli at recreational beaches in Wisconsin. The Science of the Total Environment. 404(1):10-17.

- Fahnenstiel G. L., Millie, D. F., Dyble, J., Litaker, R. W., Tester, P. A., McCormick, M. J. Rediske R., and D. Klarer. 2008. Microcystin concentrations and cell quotas in Saginaw Bay, Lake Huron. Aquatic Ecosystems Health and Management. 11(2):190 195.
- Graham, L. E. 1982. Cytology, ultrastructure, taxonomy, and phylogenetic relationships of Great Lakes filamentous algae. J. Great Lakes Res. 8:54-60.
- Hecky, R. E., R. E. H. Smith, D. R. Barton, S. J. Guildford, W. D. Taylor, M. N. Charlton, and T. Howell. 2004. The nearshore phosphorus shunt: a consequence of ecosystem engineering by dreissenids in the Laurentia Great Lakes. Can. J. Fish Aquat. Sci. 61:1285-1293.
- Herbst, R.P. 1969. Ecological factors and the distribution of *Cladophora glomerata* in the Great Lakes. Am. Midl. Nat. 82:90–98
- Higgins, S. N., E. T. Howell, R. E. Hecky, S. J. Guildford, and R.E.H. Smith. 2005. The wall of green: The status of *Cladophora glomerata* on the northern shores of Lake Erie's eastern basin, 1995-2002. J. Gt. Lakes Res. 31: 547-563.
- Hydrolab 1998. DataSonde 4a User's Manual. Hydrolab Corporation. Austin TX.
- Ishii, S., T. Yan, D.A. Shively, M.N. Byappanahalli, R.L. Whitman and M.J. Sadowsky. 2006. *Cladophora* (Chlorophyta) spp. harbor human bacterial pathogens in nearshore water of Lake Michigan, Appl. Environ. Microbiol. 72 (7):4545–4553.
- Joska, A. M., and J. J. Bolton. 1996. Filamentous freshwater macroalgae in South Africa a literature review and perspective on the development and control of weed problems. Hydrobiologia 340: 295-300.
- Keilty, T. and Woller, M. 2004. Final Report prepared for the USEPA-GLNPO by the Leelanau Watershed Council, Leelanau Conservancy.
- Landsberg JH. 2002. The effects of harmful algal blooms on aquatic organisms. Rev Fish Sci 10:113-390
- Lawrence, J.F., Niedzwiadek, B., Menard, C., Lau, B.P.-Y., Kuiper-Goodman, T., Carbone, S.; Holmes, C. 2001. Comparison of HPLC-mass spectrometry, ELISA and phosphatase assay for the determination of microcystins in blue-green algae products. J. AOAC Int. 84:1035-1044.
- MDNRE 2006. Water Quality Monitoring of Saginaw and Grand Traverse Bays. Michigan Department of Natural Resources and Environment. Lansing, MI. MI/DEQ/WB-06/096.
- Millie, D., G. Fahnenstiel, J. Dyble, R. Litaker, P. Tester, M. McCormick, R. Rediske, and D. Klarer. 2008. Influence of environmental conditions on summer cyanobacterial abundance in Saginaw Bay, Lake Huron. Aquatic Ecosystems Health and Management. 11(2):196–205.

- Mills, E. L., J. M. Casselman, R. Dermott, J. D. Fitzsimons, G. Gal, K. T. Holeck, J. A. Hoyle, O.E. Johannsson, B. F. Lantry, J. C. Makarewicz, E. S. Millard, I. F. Munawar, M. Munawar, R. O'Gorman, R. W. Owens, L. G. Rudstam, T. Schaner, and T. J. Stewart. 2003. Lake Ontario: Food web dynamics in a changing ecosystem (1970-2000). Can. J. Fish. Aquat. Sci. 60: 471–490.
- Neil, J. H., and M. B. Jackson. 1982. Monitoring *Cladophora* growth conditions and the effect of phosphorus additions at a shoreline site in northeastern Lake Erie. J. Great Lakes Res. 8:30-34.
- Olapade, O. A., M. M. Depas, E. T. Jensen, and S. L. McLellan. 2006. Microbial communities and fecal indicator bacteria associated with *Cladophora* mats on beach sites along Lake Michigan shores. Appl. Environ. Microbiol. 72:1932-1938.
- Painter, D. S., and G. Kamaitis. 1987. Reduction of *Cladophora* biomass and tissue phosphorus in Lake Ontario, 1972-1983. Can J. Fish. Aquat. Sci. 44: 2212-2215.
- Stevenson, R.J., S.T. Rier, C.M. Riseng, R.E. Shultz, and M.J. Wiley. 2006. Comparing effects of nutrients on algal blooms in streams in two regions with different disturbance regimes and with application for developing nutrient criteria. Hydrobiologia 561:149–165.
- Taft, C. E. 1975. History of *Cladophora* in the Great Lakes. *In* H. Shear and D. E. Konasewich [eds.], *Cladophora* in the Great Lakes. International Joint Commission.
- The Watershed Center 2006. Tributary Monitoring Data. The Watershed Center. Traverse City, MI.
- Vanderploeg H.A., Liebig J.R., Carmichael W.W., Agy M.A., Johengen T.H., Fahnenstiel G.L. & Nalepa T.F. 2001. Zebra mussel (Dreissena polymorpha) selective filtration promoted toxic Microcystis blooms in Saginaw Bay (Lake Huron) and Lake Erie. Can. J. Fish Aquat. Sci. 58, 1208–1221.
- Whitman, R. L., D. A. Shively, H. Pawlik, M. B. Nevers, and M. N. Byappanahalli. 2003. Occurrence of *Escherichia coli* and enterococci in *Cladophora* (Chlorophyta) in nearshore water and beach sand of Lake Michigan. Appl. Environ. Microbiol. 69:4714-4719.
- Wilson, K. A., E. T. Howell, and D. A. Jackson. 2006. Replacement of Zebra Mussels by Quagga Mussels in the Canadian nearshore of Lake Ontario: Distribution and correlations with substrate, Round Goby abundance and upwelling frequency. J. Gt. Lakes Res. 32:11-28.
- Zhu, B., D. G. Fitzgerald, C. M. Mayer, L. G. Rudstam, and E. L. Mills. 2006. Alteration of ecosystem function by Zebra Mussels in Oneida Lake: Impacts on submerged macrophytes. Ecosystems 9: 1017-1028.