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Escherichia coli and Enterococci at Beaches in the Grand Traverse Bay, Lake Michigan: Sources, Characteristics, and Environmental Pathways

SHERIDAN K. HAACK,*,† LISA R. FOGARTY,† AND CHRISTOPHER WRIGHT‡,§

U.S. Geological Survey, 6520 Mercantile Way, Suite 5, Lansing, Michigan, 48911, and The Watershed Center, Grand Traverse Bay, 232 East Front Street, Traverse City, Michigan 49684

This study quantified *Escherichia coli* (EC) and enterococci (ENT) in beach waters and dominant source materials, correlated these with ambient conditions, and determined selected EC genotypes and ENT phenotypes. Bathing-water ENT criteria were exceeded more frequently than EC criteria, providing conflicting interpretations of water quality. Dominant sources of EC and ENT were bird feces (10^8/d/bird), storm drains (10^7/d), and river water (10^{11}/d); beach sands, shallow groundwater and detritus were additional sources. Beach-water EC genotypes and ENT phenotypes formed clusters with those from all source types, reflecting diffuse inputs. Some ENT isolates had phenotypes similar to those of human pathogens and/or exhibited high-level resistance to human-use antibiotics. EC and ENT concentrations were influenced by collection time and wind direction. There was a 48−72-h lag between rainfall and elevated EC concentrations at three southern shoreline beaches, but no such lag at western and eastern shoreline beaches, reflecting the influence of beach orientation with respect to cyclic (3−5 d) summer weather patterns. In addition to local contamination sources and processes, conceptual or predictive models of Great Lakes beach water quality should consider regional weather patterns, lake hydrodynamics, and the influence of monitoring method variables (time of day, frequency).

Introduction

Recreational waters are susceptible to a variety of sources of microbiological pollution (*1*). In 1986, the U.S. Environmental Protection Agency (U.S. EPA) published numerical criteria for *Escherichia coli* (EC) and enterococci (ENT) bacteria for fresh recreational waters after determining an epidemiological linkage between gastrointestinal illness in swimmers and increasing numbers of EC and ENT in such waters (*2*). Additional studies in coastal waters have confirmed these findings (*3*, *4*). Nevertheless, monitoring of the microbiological quality of U.S. recreational waters has been inconsistently applied, and many states have not adopted these criteria (*5*).

In October 2000, the U.S. Congress passed the Beaches Environmental and Coastal Health (BEACH) Act, which requires that states with marine or Great Lakes coastal recreational waters adopt (by April 2004) water quality standards based on published U.S. EPA criteria and encourages monitoring and public notification programs through a grants program (*6*, *7*). Numbers of ENT are rarely reported for fresh recreational waters, even where monitoring programs have been established (*5*). Therefore, little information exists that specifically compares EC and ENT numbers in the same freshwater setting (*8*). As municipal, county, and state agencies develop beach-monitoring programs in response to the new legislation, new and more detailed information concerning sources, loadings, and effects of environmental factors on EC and ENT and on the characteristics of ENT in Great Lakes waters will be required.

From April to August 2000, the U.S. Geological Survey conducted a pilot study of microbiological water quality at popular beaches in the Grand Traverse Bay near Traverse City, MI. The pilot study was followed by a trial monitoring program for major Grand Traverse Bay and nearby Lake Michigan beaches conducted by the Watershed Center of Grand Traverse Bay and the Grand Traverse County and Benzie-Leelanau District Health Departments. Together, the pilot study and trial monitoring program addressed several basic requirements for establishing a monitoring and public notification program under the BEACH Act (*6*), including (i) the assessment of the proximity of point and nonpoint sources of pollution; (ii) the effects of storm events on beach-water quality; and (iii) the evaluation of monitoring methods and assessment procedures in light of site-specific variables. The beaches studied are typical of many popular Great Lakes beaches located outside large urban areas. They receive intense recreational use in the summer months from local residents and tourists, combined sewer overflows or large wastewater streams are rare, and monitoring of recreational water quality has been infrequent. Processes influencing recreational water quality at such beaches remain poorly understood.

Goals of the pilot study were to determine the nature and significance of and environmental processes contributing to EC and ENT contamination at representative beaches. The goals of the trial monitoring program were to evaluate standardized monitoring approaches, incorporating State of Michigan requirements for number and frequency of EC samples, and to extend the data set of EC concentrations and associated environmental variables over more of the swimming season and to more beaches. Overall objectives were to (i) quantify EC and ENT in dominant source materials and recreational waters; (ii) characterize selected source isolates using genomic (EC) or biochemical (ENT) profiling; (iii) identify associations between numbers of these two indicator bacteria groups and ambient conditions; (iv) identify processes that influence spatiotemporal variability of indicator bacteria at these beaches; and (v) evaluate standardized monitoring approaches in light of site-specific knowledge about sources and environmental processes. This paper offers insights into factors affecting recreational water quality at similar Great Lakes beaches and provides information useful in the design of recreational water-quality monitoring and public notification programs.

* Corresponding author phone: (517)887-8909; fax: (517)887-8937; e-mail: skhaack@usgs.gov.

† U.S. Geological Survey.

‡ The Watershed Center.

§ Present address: Michigan State University, Department of Agricultural Economics, 202 Agriculture Hall, East Lansing, MI, 48824.

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Experimental Section

Water Quality Standards. For designated freshwater beaches, the U.S. EPA steady-state geometric mean criterion is 126 EC or 33 ENT/100 mL, and the single-sample maximum criterion is 235 EC or 61 ENT/100 mL (*2*). Michigan does not require beach monitoring, but Michigan Water Quality Standards contain numerical criteria for EC to be met when monitoring is conducted. These criteria are a 30-d geometric mean (from 5 or more sampling dates) not to exceed 130 EC/100 mL or a single sample (defined as the geometric mean of triplicate site samples) not to exceed 300 EC/100 mL. Michigan has published no criteria for ENT.

Study Area. The study area is shown in Figure S-1 (Supporting Information). Traverse City maintains separated sewer and stormwater drainage systems. Wastewater enters the Boardman River (draining 733 km^2; Figure S-1) following final UV disinfection. Upstream of the wastewater treatment plant (WWTP), forest (62%) and agricultural (24%) land uses predominate, but from the WWTP to the mouth, land use is urban and the river is channelized with approximately 30 storm drains, catch basins, or other outfalls in this reach, many of which flow regardless of rainfall. Storm drains (0.76–1.1 m diameter corrugated metal or concrete pipes) are located at West End Beach (WB) and Bryant Park Beach (BB) but not at Clinch Park Beach (CB) (Figure S-1). In 2000–2001, Lake Michigan water levels were 0.6 m below average (*9*), and only the BB drain opened under water, approximately 15 m off shore. Yuba Beach (YB) and Sutton's Bay Beach (SUT) are on the east and west shores (respectively) of Grand Traverse Bay. Yuba Beach may be affected by flow from Yuba Creek, which drains primarily undeveloped land. Sutton's Bay Beach is on the northern edge of a small municipality and is susceptible to inputs similar to those at CB.

Sample Collection. In the pilot study, beach (swimming) water, storm-drain water, river water, groundwater, beach sediments, floating detritus, and feces from gulls, ducks, pigeons, and geese were analyzed for EC and ENT densities. In the monitoring program, as required by the State of Michigan, only beach water was analyzed and only for EC densities. Beach (swimming) water was collected at ankle (0.15 m) or knee (0.5 m) water depths (pilot study) or chest height (1.3 m depth, monitoring program). Sediments were collected in the swash zone (wave-wetted margin) or in ankle- or knee-deep water. Water sample collection and processing procedures followed ref *10*. Weather data were from the National Oceanic and Atmospheric Administration Traverse City Airport station. Total suspended solids (TSS) analysis followed ref *11*. Specific conductance (SC), pH, and temperature of water were recorded in the field with hand-held meters. In the pilot study, samples were collected on arbitrary dates, and a variety of types of samples were collected on each date. In the monitoring program, following requirements of the State of Michigan, beaches were sampled 5 times per 30-d interval, from three locations at each beach. The geometric mean of these three samples was calculated for each sampling date, and the moving 30-d geometric mean was recorded throughout the sampling period (May 15–September 19, 2001).

Bacteria Enumeration. In the pilot study, total coliform bacteria (TC), EC, and ENT were quantified by membrane filtration (*10*, *11*): TC on mENDO agar LES, EC on NA-MUG agar, and ENT on mEI agar (DIFCO Laboratories, Detroit, MI). EC and ENT were confirmed as described in ref *10*. In the monitoring program, sample collection and analyses were conducted by SOS Analytical, Inc. (Traverse City, MI), and EC were tested using QuantiTray/2000 (IDEXX Laboratories, Inc., Westbrook, ME).

Pilot Study Source Survey. Solid samples were weighed, suspended in phosphate-buffered saline, diluted in series, and thereafter treated as water samples for microbiological analysis. Beach sediments were predominantly coarse sands. Sediments were collected by grinding the open mouth of a sterile bottle into the sediments. The effect of sediments on numbers of indicator bacteria in the water column was evaluated by sampling overlying water before and after rapidly and repeatedly suspending sediments to a depth of 15 cm over an area of 30 cm^2. Samples of water containing detritus were analyzed as typical beach-water samples after shaking and allowing the largest particles to settle. Fecal samples were collected with a sterile spatula from gull, goose, pigeon, and duck feces (n = 16) just after defecation.

Groundwater beneath the lake bed was collected about 10 m from shore in knee-deep lake water, through a 5-cm-diameter, 1.8-m-long steel drive-point piezometer of in-house construction. The piezometer was driven to a depth of 0.7 m into the lake bed and then a 30-cm-diameter PVC tube, 1 m in length, with a hole drilled in the side at 30 cm from the bottom was placed over the piezometer to create a stilling well. To establish the vertical hydraulic gradient, water level in the piezometer (groundwater) was measured relative to that in the surrounding stilling well (lake level) with a water-activated electronic tape. After being purged for 5 min, water was sampled from the piezometer with a peristaltic pump fitted with sterile tubing.

River and storm-drain water was sampled throughout the pilot study. Effluent from the WWTP was collected twice, just prior to UV treatment. On October 8, 2000, 16.3 mm rain fell between 1130 and 1500 h. CB beach water, WB parking-lot runoff, WB storm-drain water, and river water samples were collected before, during, and for 3 d following the storm.

Characterization of EC and ENT in Source Materials and Beach Water. Genomic profiles of EC were obtained using rep-PCR with the primers REP 1R and REP 2I (*12*, *13*; Genosys Biotechnologies, The Woodlands, TX). *E. coli* ATCC 25922 was included as a control. *Enterococcus* phenotypes were defined using multiple physiologic assays (API Rapid ID 32 Strep, bioMérieux, Hazelwood, MO) as well as colony color and hemolysis on Columbia sheep blood agar (BBL Becton Dickinson). *Ent. faecalis* ATCC 19433 was used as control. Resistance of selected ENT to the antibiotics tetracycline, ampicillin, streptomycin, gentamicin, and vancomycin was determined using the Etest (AB Biodisk, Piscataway, NJ). The choice of these five antibiotics (commonly used to treat enterococcal infections in humans) and definitions of resistance to each antibiotic were based on National Committee for Clinical Laboratory Standards criteria (*14*).

Statistics. Bacterial numbers were transformed [log(x + 1)]. Statistical analyses (correlations and t-tests; SigmaStat, Jandel Scientific, San Rafael, CA) were performed on the transformed data. Statistical procedures for cluster analysis of EC genotypes and ENT phenotypes are described in Figures S-2 and S-3 (Supporting Information).

Results and Discussion

Overview of Recreational Water Quality. *Pilot Study.* ENT-based recreational water-quality criteria were exceeded more frequently than EC-based. A total of 42 samples of beach water were collected (Table 1) at ankle (n = 23) or knee depth (n = 19). There was no significant difference (t-test) in concentrations of either EC or ENT at ankle versus knee depth. The greater number of ENT exceedances did not necessarily result from larger concentrations of ENT than EC. The median EC to ENT ratio was 1.44 for all beach water samples. In the four beach-water samples exceeding the single-sample EC criterion (Table 1), this ratio ranged from 4.6 to 46.0, possibly indicating a source of EC-dominated contamination. In contrast, this ratio ranged from 0.1 to 11.3 with a median of

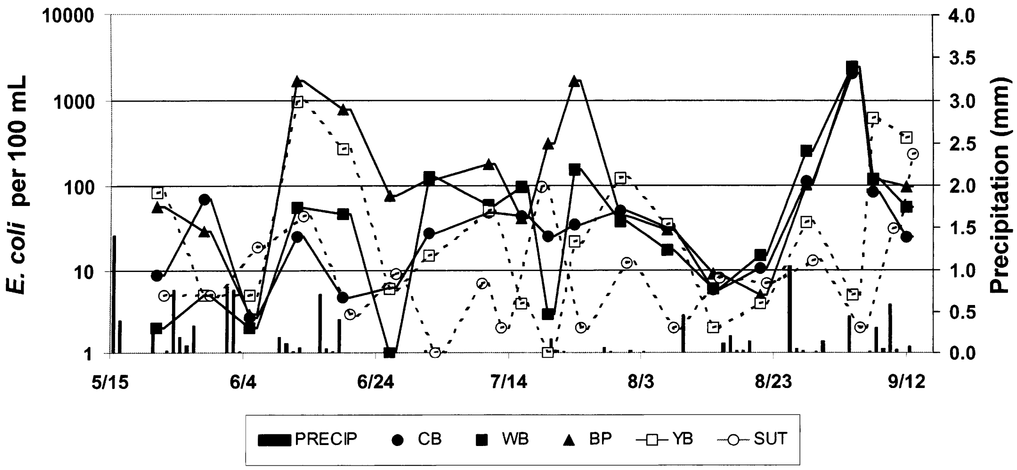

FIGURE 1. Precipitation and EC concentrations in beach water for May 15–September 19, 2001. CB, Clinch Beach; WB, West Beach; BB, Bryant Park Beach; YB, Yuba Beach; SUT, Sutton's Bay Beach.

TABLE 1. Exceedances of U.S. EPA Recreational Water Quality Criteria at Beaches in the Grand Traverse Bay[a]

	pilot study					monitoring program		
beach	*n*	EC single	EC steady[b]	ENT single	ENT steady[b]	n	EC single	EC steady[b]
CB	28	3	no	6	yes	20	2	1
WB	12	0	no	4	yes	20	2	2
BB	2	1	yes	1	yes	20	5	5
YB	ns					20	4	0
SUT	ns					20	1	0

[a] *n*, number of samples. Each monitoring program single sample represents the geometric mean of three beach water samples collected at the same date and time. Single, single-sample criterion exceedance. Steady, steady-state criterion exceedance. ns, not sampled. [b] Only one steady-state calculation was performed for all samples collected during the pilot study. In the monitoring program, the running 30-d geometric mean was calculated.

TABLE 2. Bacteria Densities in Beach Water, Water with Suspended Sediment or Detritus, and Groundwater[a]

	no.[b]/100 mL		
	total coliforms	EC	ENT
Sediment Suspension			
beach water (6/23-CB)	32	4	10
water after sediment suspension	29	1	nt
beach water (8/10-CB)	40	10	2
water after sediment suspension	70	20	5
Detritus			
beach water (8/8-CB)	312	156	23
water + detritus	570	320	100
beach water (8/9-CB)	120	90	58
water + detritus	3100	1800	30
Groundwater			
beach water (8/8-CB)	653	106	27
groundwater	360	120	71
beach water (8/9-WB)	71	35	39
groundwater	210	100	119
beach water (8/9-CB)	180	95	58
groundwater	650	320	182
beach water (8/10-CB)	49	14	3
groundwater	520	30	16

[a] CB, Clinch Beach; WB, West Beach. nt, not tested. [b] Geometric mean of beach water samples taken at same location, date, and time as that of test samples.

1.0 in the 11 beach-water samples exceeding the single-sample ENT criterion. A comparison study of fecal coliforms and ENT at New York marine and freshwater beaches (*15*) found that the ENT standard would result in increased beach closures, especially in freshwater lakes, ponds, and marine harbors or bays—settings comparable to Grand Traverse Bay. Likewise, in a recent study of EC and ENT at Lake Michigan beaches in Wisconsin, ENT single-sample and geometric mean criteria were exceeded more frequently than were EC criteria (*8*). Results such as these, in which EC and ENT indicate different recreational water quality for the same samples or beaches, pose a dilemma for beach managers and indicate that further research on the nature, sources, and public health significance of these two indicators is warranted.

Monitoring Program. Recreational water quality EC criteria also were exceeded during the 2001 monitoring program (Table 1, Figure 1). The monitoring program revealed that the three Traverse City beaches (CB, WB, and BB) tended to follow similar trends in EC concentrations (Figure 1), while YB and SUT, on the east and west shores, respectively, did not clearly follow the same trend. As in the pilot study, among all beaches, BB was most susceptible to EC contamination (Table 1), possibly due to the large storm drain that opens directly offshore.

Pilot Study Source Survey. *Sediments and Detritus.* The geometric mean numbers of TC, EC, and ENT/g wet weight of sediment were 3, 2, and 2, respectively ($n = 21$). There was little effect of sediment suspension on indicator bacteria counts in the overlying water (Table 2). Coarse sands such as those at Traverse City beaches generally have low numbers of indicator bacteria and little effect on delivery of bacteria to water (*16*–*18*).

Detrital material, especially decaying vegetation, has been cited as a significant source of FC (*18*) or ENT (*19*) at beaches. Detritus at Traverse City beaches variously consisted of small (<2 cm) fragments of wood, leaves from terrestrial or aquatic plants, insects or crustaceans, feathers, dead fish, masses of dead or living but detached zebra mussels (*Dreissena* sp.), and occasional garbage. Both water + detritus samples (Table 2) exhibited much larger numbers of TC and EC than the corresponding beach-water samples and very high TSS values in the supernatant. Fine-grained sediments have been implicated as a source of EC and ENT (*17*, *20*) and of some bacterial pathogens (*21*, *22*). Indicator bacteria may accumulate in storm-drain and river sediments and be delivered with high TSS waters during runoff events (*23*, *24*). Particles in water protect bacteria from the effects of UV light or predators (*17*). Apparently, sands at Traverse City beaches

TABLE 3. Bacteria Contributions of Surface Runoff and Boardman River—Pilot Study

	flow rate (m^3/s)	TSS (mg/L)	no./100 mL (loading rate) TC	EC	ENT
			During Dry Conditions		
West Beach drain[a]	0.00053	22.8	277 (1.26×10^8/d)	127 (5.81×10^7/d)	200 (9.16×10^7/d)
Boardman River[b]	5.8	3.3	363 (1.81×10^{12}/d)	137 (6.86×10^{11}/d)	135 (6.76×10^{11}/d)
			During Storm of August 8, 2000		
West Beach drain	0.0018	233.0	4000 (2.59×10^8/h)	1600 (1.03×10^8/h)	14500 (9.39×10^8/h)
parking lot runoff	nd[c]	88.0	5700	5000	10200
Boardman River	4.47	8.0	7500 (1.21×10^{12}/h)	1000 (1.61×10^{11}/h)	3300 (5.31×10^{11}/h)

[a] Five observations for West Beach drain. [b] Six observations for Boardman River. [c] nd, not determined.

contribute relatively few bacteria to beach waters, but detritus or fine-grained materials (likely delivered during runoff as described below) may have a significant effect on recreational water quality.

Groundwater. The interaction between surface and subsurface water at beaches is poorly understood and rarely studied in freshwater settings (*25*). Groundwater contributions to beach bacteria numbers have rarely been investigated (*18*, *26*). In eight observations, groundwater exhibited an upward vertical hydraulic gradient at CB and WB. Groundwater typically exhibited a 2−10× greater concentration of all indicator bacteria than corresponding beach water (Table 2). At these specific beaches, leaking sewer lines or storm drains might be a source, as implicated in refs *18* and *26*; however, surface runoff infiltration and wave or seiche washing of beach sands might also deliver bacteria to shallow beach groundwater (*25*). The contribution of groundwater to the Grand Traverse Bay through direct flow is small (7%) as compared to the amount delivered as base flow in tributaries (*27*). Nevertheless, high concentrations of indicator bacteria in groundwater with an upward hydraulic gradient might have immediate and measurable effects on water samples collected in shallow lake water, especially during calm conditions (morning, SW winds; see below) when local mixing with lake waters is reduced.

Birds. Gulls (typically 30−50 per beach) and ducks (<15 per beach) occurred at all beaches, but pigeons and geese were infrequent during the swimming season. All birds were numerous along urban sidewalks, grassy areas, and river banks. The ratio of EC to ENT was similar in fecal samples from all bird types, but goose feces contained the lowest numbers of all bacteria types and pigeon feces contained the highest. Numbers/g (wet weight) averaged 2.9×10^6 EC [n =16, range: 4.2×10^3 (goose) to 1.3×10^{10} (pigeon)] and 3.1×10^6 ENT [n =13, range: 5.0×10^2 (goose) to 1.4×10^{10} (pigeon)]. Gull and duck feces averaged 1.4×10^7 EC and 5.0×10^7 ENT/g. The average wet weight of feces excreted by different gull species ranges from 11.2 to 24.9 g/day (*28*). Assuming a similar amount for ducks, this would result in a range of daily loads from one gull or duck of $1.6−3.4 \times 10^8$ EC and $5.6 \times 10^8−1.3 \times 10^9$ ENT. These estimates are within the ranges reported by other coastal studies (*18*, *19*). Extrapolating from these calculations, the daily loading from 50 birds could result in a range of concentrations between 775 and 1720 EC and 2810−6250 ENT/100 mL for a swimming area 100 m long, 10 m offshore, and 1 m deep. Because bird feces are delivered to these beaches by multiple pathways—direct deposition in the water, washing from the swash zone sands, and parking lot, sidewalk, storm drain, and river runoff during storms—bird feces are likely one significant source of bacterial contamination to these beaches.

Storm Drains and the River. Storm drains and the Boardman River contributed large numbers of EC and ENT to the bay, even during nonrunoff conditions (<1 mm rainfall in the preceding 24 h; Table 3). The WB drain average SC was 458 μS/cm (average beach-water SC, 350 μS/cm), TSS was 22.8 mg/L (beach water, 8.5 mg/L), and temperature was 13 °C (beach water, 19 °C). The temperature differential suggests that groundwater contributes to drain flow during dry conditions. On two dates, samples collected between the location of the WWTP and the mouth of the river (Figure S-1) indicated that TC and EC were higher at the river mouth (for the two dates: 370 and 380 TC; 80 and 70 EC) than at 8th Street (90 and 80 TC; 0 and 10 EC) or Union Street (140 and 46 TC; 0 and 7 EC). Bacteria numbers in the WWTP effluent collected prior to final UV treatment were 4.1×10^3 TC, 2.5×10^3 EC, and <100 ENT/100 mL. Numbers would presumably be even lower in the final effluent. Low bacteria concentrations in the river nearest the WWTP suggest that the WWTP is not a significant source of bacterial contamination during typical conditions. However, bacteria injured during the treatment process and therefore unable to grow on typical media might become resuscitated by the time they reach the mouth of the river (*29*). Alternatively, increasing river bacteria concentrations downstream toward the river mouth could be due to inputs from the numerous storm and catch basin outfalls in this reach.

The rainstorm of August 8, 2000, caused an immediate increase in bacterial numbers in storm drain and river runoff (Table 3, Figure 2). During the storm, WB drainwater SC decreased (214 μS/cm) and temperature increased (18 °C) from typical values, reflecting the surface-runoff source. In addition, TSS increased (Table 3), probably contributing to the increased delivery of bacteria (*23*, *24*). Total coliforms and enterococci are broad groupings of related bacteria that include both fecal bacteria and bacteria native to the environment. In contrast, except for tropical environments (*30*), *E. coli* are not generally considered to be present in the environment except as fecal contaminants. EC numbers may most accurately reflect the fecal component of storm-drain or river indicator bacteria, while TC genera and ENT species in the river and storm drains could vary under different hydrologic or environmental conditions. The increase in river bacteria numbers (measured at the mouth) immediately during the storm and the decline in numbers on following days even though river discharge increased slightly on the second day (Figure 2) might indicate that the source of these bacteria is near the river mouth, as opposed to a distant watershed source, as we also noted during dry conditions. Until 74 h following the August 8, 2000 storm, there was little effect on bacterial concentrations in CB water (Figure 2). This observation will be discussed below.

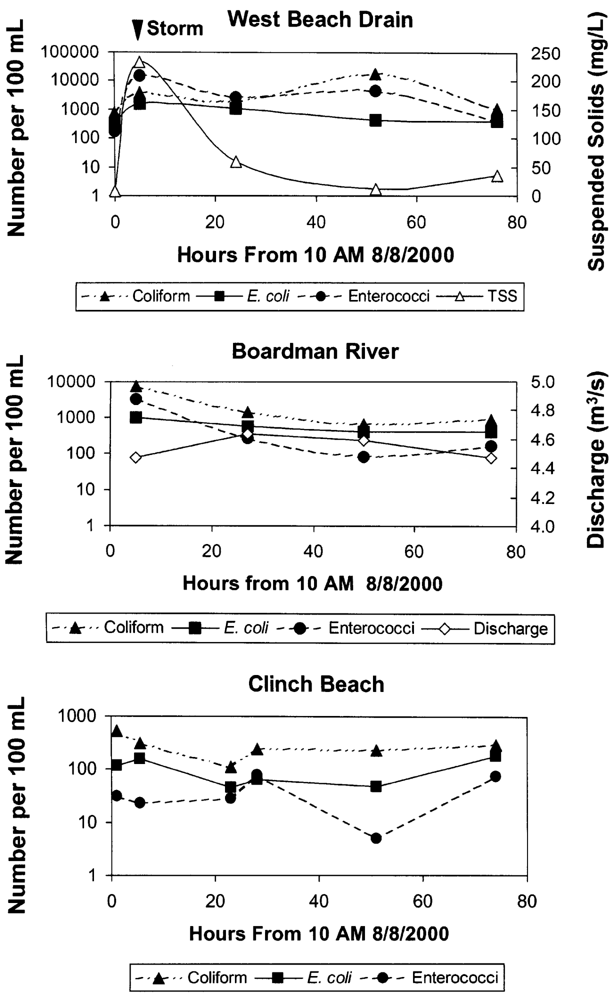

FIGURE 2. Indicator bacteria concentrations and environmental parameters measured in river, storm-drain, and beach water before, during, and after a rainstorm on August 8, 2000.

Association with Ambient Conditions and Environmental Processes. In the pilot study, all indicator bacteria concentrations were positively correlated with wind speed and with each other, suggesting that common factors influenced their occurrence (Table 4). TC and EC concentrations were most highly correlated with each other. Pilot study TC, EC, and ENT were not correlated with rainfall; however, only one sampling date during the dry summer of 2000 followed a significant rainfall in the preceding 24, 48, or 72 h. Median TC, EC, and ENT concentrations all tended to be greater in the morning (0900–1200 h) than in the afternoon (1230–1630 h) (Figure 3). However, the most extreme (highest and lowest) concentrations of TC, EC, and ENT occurred in the afternoon (Figure 3). Several prior studies have reported the antibacterial effects of UV light over the course of the day (*31*–*33*), which may contribute to the lower mean or 50th percentile concentrations for afternoon samples. However, lake breezes develop on most summer days, and wind speeds are greatest in midday or afternoon with calm conditions in the AM or PM. In addition to being correlated with wind speed (Table 4), TC, EC, and ENT concentrations were also associated with wind direction (Figure 4), and there was a relation between wind direction and speed and time of day: 15/17 times the morning wind direction was from the NW and frequently (10/17 times) wind speed was low and wave height was zero. In contrast, in the afternoon, wind speed was generally higher, and zero wave height was only observed when SW winds (4/22 observations) drove water away from the beaches. Diurnal variability in wind speed and direction

TABLE 4. Correlation Coefficients for Indicator Bacteria Densities versus Ambient Environmental Conditions—Pilot Study[a]

	correlation coefficient for geometric mean no./100 mL		
factor	total coliforms	EC	ENT
water temp (°C)	−0.193	−0.256	−0.426
SC[b] (μS/cm)	−0.094	−0.085	0.058
TSS[c] (mg/L)	0.146	0.100	0.395
rain 24 h (mm)	−0.113	−0.100	−0.062
rain 48 h (mm)	−0.154	−0.159	−0.240
rain 72 h (mm)	−0.068	−0.067	−0.112
wave height (cm)	0.207	0.237	0.077
wind speed (m/s)	0.726*	0.756*	0.600*
no. of swimmers	−0.120	−0.147	0.144
no. of people	−0.141	−0.142	0.034
no. of birds	−0.003	−0.098	0.192
TC	1	0.986*	0.635*
EC	0.986*	1	0.586*
ENT	0.635*	0.586*	1

[a] An asterisk (*) indicates significant at $p < 0.05$. [b] SC, specific conductance. [c] TSS, total suspended solids.

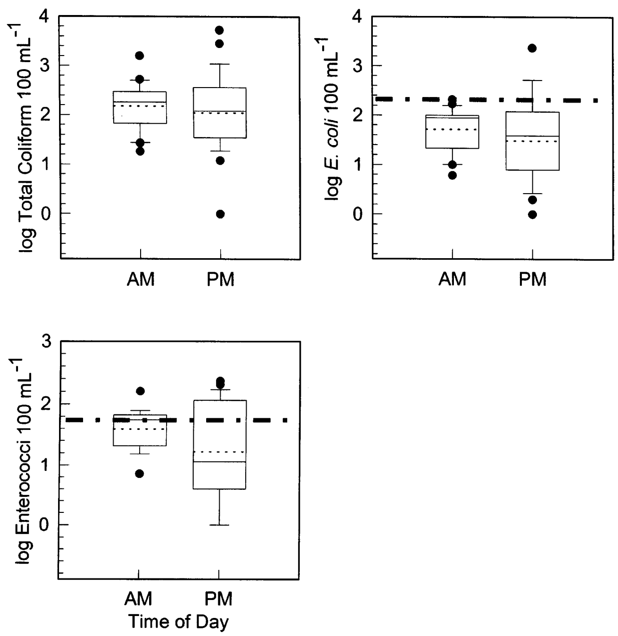

FIGURE 3. Relation between indicator bacteria concentrations in beach water and time of sampling at CB, WB, and BB. AM = 0830–1200 h; PM = 1230–1630 h. Box plots show the 90th, 75th, and 50th (solid line); mean (dotted line); and 25th and 10th percentiles of log(x + 1) transformed data. The U.S. EPA-recommended single-sample standard (dot–dash line) is also indicated for EC and ENT.

may contribute to the extreme values for TC, EC, and ENT concentrations (Figure 4). Further testing would be required to establish the independent and interactive effects on TC, EC, and ENT concentrations of sunlight, wave height, wind direction, and wind speed.

Monitoring program samples were collected in the AM (0830–1200 h) and in deeper water than in the pilot study. Antecedent rainfall was more frequent than in the pilot study, although summer rainfall in 2001 was still below average (Figure 1). As in the pilot study, EC at CB, WB, and BB were positively correlated with each other (Table 5). In contrast to the pilot study, EC were not significantly correlated with wind speed, perhaps because samples were collected in the AM when, as described above, wind speeds were generally low. Additionally, EC at CB and WB were correlated with

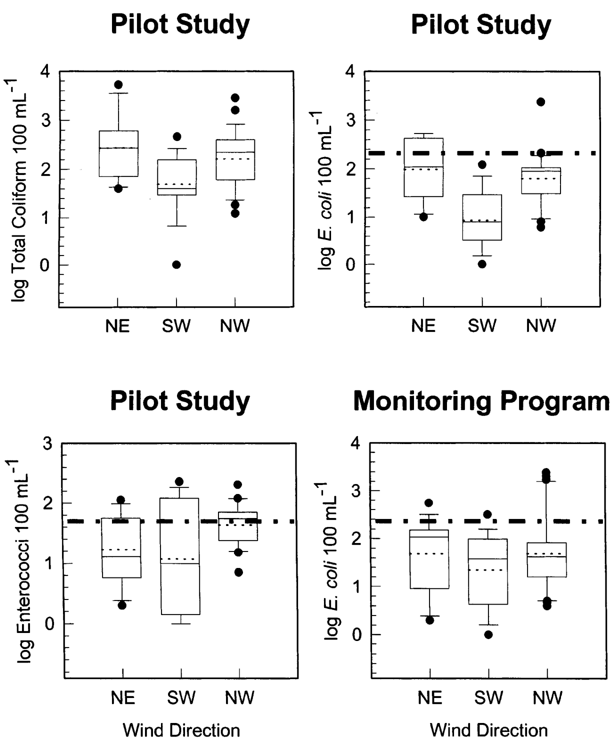

FIGURE 4. Relation between indicator bacteria numbers and wind direction at CB, WB, and BB. Box plots show the 90th, 75th, and 50th (solid line); mean (dotted line); and 25th and 10th percentiles of log(x + 1) transformed data. The U.S. EPA-recommended single-sample standard (dot−dash line) is also indicated for EC and ENT.

TABLE 5. Correlation Coefficients for EC Densities versus Ambient Environmental Conditions—Monitoring Program[a]

	correlation coefficient for geometric mean of EC/100 mL				
factor	Clinch Beach	West End Beach	Bryant Park Beach	Yuba Beach	Sutton's Bay Beach
water temp (°C)	0.173	−0.090	−0.072	−0.065	−0.080
TSS[b] (mg/L)	0.954*	0.777*	0.234	0.246	0.954*
rain 24 h (mm)	−0.106	−0.087	−0.041	0.554*	0.766*
rain 48 h (mm)	0.512*	0.533*	0.443*	0.260	0.672*
rain 72 h (mm)	0.443*	0.459*	0.406	0.302	0.524*
wave height (cm)	0.314	0.181	0.093	0.054	0.848*
wind speed (m/s)	0.125	0.205	0.275	−0.014	0.301
Clinch Beach EC	1	0.965*	0.644*	−0.133	0.094
West Beach EC	0.965*	1	0.699*	−0.109	−0.104
Bryant Park EC	0.644*	0.699*	1	0.294	−0.110
Yuba Beach EC	−0.133	−0.109	0.294	1	0.130
Sutton's Bay EC	0.094	−0.104	−0.110	0.130	1

[a] An asterisk (*) indicates significant at $p < 0.05$. [b] TSS, total suspended solids.

TSS, which was not a significant correlation in the pilot study. EC at these three beaches were correlated with rainfall in the preceding 48−72 h but not with rainfall in the preceding 24 h (Table 5). Similarly, in the pilot study, EC numbers at CB were highest 74 h after the storm of August 8, 2000 (Figure 2), in concert with (i) large amounts of detritus in the form of dead insects and feathers, (ii) relatively high waves (30 cm), and (iii) a shift in wind direction from SW on the day of the storm to NW−NE 3 d later. In Grand Traverse Bay, summer wind direction and precipitation typically follow a 3−5 d cycle of low- and high-pressure systems. Low-pressure systems bring S−SW winds (driving water away from the southern-end beaches) and rain. Following passage of the low-pressure system by 3−5 d, winds shift to NW−NE, and sunny conditions predominate. NW−NE winds would drive water down the Bay toward Traverse City, and shifts from low to high pressure might cause seiche events (*34*). The correlation between EC concentrations at CB, WB, and BB with antecedent 48−72-h rainfall may be associated with a wind- or seiche-driven circulation pattern, bringing previously delivered runoff contamination to these beaches. An interesting contrast is provided by EC concentrations at two beaches (YB and SUT) on the east and west shores of the bay (Table 5), which are oriented differently with respect to weather patterns and typical wind directions. EC concentrations at YB and SUT were (i) correlated with 24-h rainfall instead of 48−72-h rainfall; (ii) were not correlated with concentrations at CB, WB, and BB; and (iii) were highest at different times than the southern-end beaches (Figure 1).

Seiches and wind-driven circulation patterns are common to all lakes and are not restricted to bodies of water such as Grand Traverse Bay. Individual Great Lakes beaches will experience varying relative contributions from local sources (e.g., washing of bird feces from the shoreline swash zone, groundwater seepage, storm-drain contributions under dry conditions, or contributions directly to the water by birds) under varying ambient conditions. If bacteria concentrations exhibit a cyclic pattern in response to weather patterns or vary systematically under different ambient conditions (e.g., AM vs PM or calm vs turbulent water), different monitoring schedules may yield different trends in recreational water quality. Results presented here indicate that, to adequately understand sources and environmental processes influencing beach bacteria concentrations at Great Lakes beaches, consideration must be given to (i) beach orientation with respect to regional weather patterns; (ii) regional and local hydrodynamics; (iii) nature, timing, and magnitude of various local source inputs; and (iv) interactions between these factors.

Characterization of EC and ENT in Source Materials and Beach Water. To provide further insight into the nature and significance of EC and ENT contamination, we determined rep-PCR genomic profiles for 81 EC isolates (Figure S-2) and phenotypic profiles for 64 ENT isolates (Figure S-3) from beach water and dominant sources. Most isolates were acquired between August 8 and August 11, 2000. rep-PCR produced very fine discrimination between EC isolates, with different source samples (including the 12 bird samples) typically yielding one dominant and one to four additional genotypes. There were four EC genomic profile matches between different sources at the 90−100% level of similarity (Figure S-2). Beach-water EC isolates occurred in clusters with all source types, only two beach-water isolates matched at a satisfactory level of similarity with any specific source isolate (storm drain, river), and there were no patterns of EC clustering with respect to source. Likewise, beach-water ENT isolates occurred in clusters with all source types, and no beach-water ENT isolate was matched with a source ENT isolate at 100% similarity (Figure S-3). Studies that apply genomic or phenotypic profiling as the only means of source determination typically acquire hundreds of isolates from known sources, construct a source library, and then match environmental isolates to this library using multivariate statistics (*35−40*). The large number of genotypes and phenotypes found in these relatively small samples of EC and ENT isolates indicates that very large numbers of isolates would be needed to fully characterize sources or find matches to beach-water isolates. More intensive sampling of each source type would likely reveal additional EC profiles in each source. Testing of other animals for which feces may be delivered to urban stormwater (dogs, cats, rodents, raccoons) or of humans swimming in the water (*41*) might reveal additional EC or ENT types that would match those in Traverse City beach-, river-, or drainwater. In addition, if

TABLE 6. Minimum Antibiotic Concentration Inhibiting 90% of *Enterococcus* Isolates (MIC_{90}) from the Indicated Source

	MIC_{90} (μg/mL)				
source	streptomycin $>2000^a$	gentamicin $>500^a$	vancomycin $8-16^b$	tetracycline $\geq 16^c$	ampicillin $\geq 16^c$
bird	$\geq 256^d$	$\geq 1024^e$	6	$\geq 256^d$	3
effluent or runoff	$\geq 256^d$	24	3	64	4
sediment	$\geq 256^d$	32	4	96	4
river	$\geq 256^d$	16	12	64	4
beach water	$\geq 256^d$	24	6	0.75	3

[a] High-level resistance; many enterococci are inherently resistant to lower levels of this antibiotic. [b] Range for intermediate resistance requiring further testing. [c] Defined resistance level. [d] Greatest concentration tested was 256 μg/mL. [e] Greatest concentration tested was 1024 μg/mL.

large-scale circulation patterns bring contaminants from other locations to Traverse City beaches, then additional sources (e.g., cattle, horses, deer, turkeys, septic systems, small community wastewater streams) in the Grand Traverse Bay watershed should be sampled. Nevertheless, the broad association of EC and ENT beach-water isolates with those from all types of sources in multiple clusters is consistent with nonpoint pollution sources and with our observations that beach-, river-, and drainwater all are susceptible to the same direct sources of contamination and are connected through hydrologic processes.

ENT were responsible for the largest number of exceedances of recreational water-quality criteria in the pilot study. ENT exhibited variation in phenotypic and antibiotic resistance profiles with source (Table 6, Figure S-3)—features that are useful for source determination studies (*38*−*40*). ENT similar to *Ent. faecium* were found in storm-drain and parking lot runoff, river water, and the WWTP but not in bird feces. ENT similar to *Ent. hirae* or *durans* were found primarily in bird feces and sediment, and ENT similar to *Ent. faecalis* were found only in bird feces and beach water. Isolates from birds were more likely to be resistant to both streptomycin and tetracycline at the highest tested concentration (256 μg/mL; Table 6), and two of three isolates exhibiting high-level (>1000 μg/mL) gentamicin resistance were from birds.

These data also suggest environmental ENT in Grand Traverse Bay beach waters and source materials may have human medical significance. *Ent. faecalis* and *faecium* have emerged in the past decade as leading causes of hospital-acquired infections, in which their intrinsic and acquired antibiotic resistance presents significant treatment challenges (*42*, *43*). ENT are abundant in soil and on plant surfaces where new genotypes of typical fecal indicators (*Ent. faecium* and *Ent. faecalis*) have been identified (*44*). The potential health risk of these environmentally occurring *Ent. faecium* and *Ent. faecalis* remains unknown. The antibiotics we chose for analysis are those used most frequently in the treatment of enterococcal infections. The prevalence of ENT in these representative Great Lakes beach waters, and their resistance patterns with respect to medically relevant antibiotics warrant further study.

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Supporting Information Available

Figures depicting the study area (S-1), cluster analysis of *E. coli* genomic profiles (S-2), and cluster analysis of enterococci phenotypic profiles (S-3). This material is available free of charge via the Internet at http://pubs.acs.org.

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