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There are about 20,000 wastewater treatment plants in the United States. These plants discharge about 50 trillion gallons of wastewater daily into the nation's surface waters (Dorfman, 2004). Most wastewater contains human feces, which are a potential source of microbial pathogens. Pathogens that may be found in the sewage include bacteria, viruses, and protozoa. Table 1 lists some of the pathogens that have been isolated from wastewaters. All of these microorganisms are transmitted via the fecal-oral route; therefore, if wastewater is discharged to surface waters, they pose a health risk to anyone who comes in contact with the water or who consumes food harvested from the water. The potential risks that are associated with wastewater make disposal and control of wastewater a significant public health issue.

RECREATIONAL WATERS AND HUMAN HEALTH

The human populations most at risk of being exposed to these wastewaters are recreational users who swim or participate in other water-related activities, such as surfing, sailboarding, and snorkeling. In the years 1999–2000, more than 25 percent of the population 16 years and older participated in coastal water swimming (Leeworthy and Wiley, 2001). This represents a swimming population of 52.6 million individuals, and an estimated 758 million days of swimming activity. Similarly, it is estimated that 10.5 million individuals undertook 92 million days of snorkeling; 2.8 million individuals spent an estimated 23 million days of scuba diving; 33 million surfers participated in 76 million days of surfing; and somewhat less than 800,000 wind surfers participated in 5.8 million days of windsurfing. All of these modes of water exposures pose potential health risks to recreational users of coastal waters.

The health effects associated with water activities, however, are not easily detected. One approach to establishing the linkage between illnesses in swimmers and swimming activity is to use surveillance systems to detect outbreaks of disease when they occur. Outbreaks of disease are usually detected when individuals who become ill after engaging in the same activity are identified by physicians as having a common linkage to the same exposure factor. Between 1971 and 2002, only two outbreaks of disease associated with swimming in marine waters were reported (Craun et al., 2005). During that same period, 257 outbreaks of disease were reported from freshwater and treated water (swimming pool) environments. The reason for the small number of marine water outbreaks is not known, but one can speculate that marine waters may be of higher quality than those of freshwaters; or that many freshwater

outbreaks are due to swimmer to swimmer transmission through the water; or because swimmers at ocean beaches tend to travel further, marine water outbreaks may be less well reported.

The other approach to identifying health effects associated with recreational waters is through epidemiological studies in which a large group of individuals who are exposed to risk factors are compared to a control group after a period of time to determine if there are differences in their respective health status. Between 1972 and 1978, the United States Environmental Protection Agency (US EPA) carried out a series of epidemiological studies at marine bathing beaches (Cabelli, 1983). The objective of those studies was to determine the relationship between domestic sewage-contaminated beach water and swimming-associated illness, and further, to determine the best way to measure the contamination. The studies were conducted at beaches where the water quality was barely acceptable and at beaches where the water quality was good. Swimmers and non-swimmers were given an initial interview, and then, seven to ten days later, a follow-up interview was given in order to determine their health status in the intervening time period. The difference in illness rates of these two groups was attributed to the level of fecal contaminants in the water at the barely acceptable beach, as

Table 1. Pathogens Associated with Wastewaters

Organism	Concentration in Wastewater (per 100 mL)	Disease		
Bacteria				
Campylobacter	3,700-100,000	Gastroenteritis		
Pathogenic E. coli	30,000-10,000,000	Gastroenteritis		
Salmonella	0.2-11,000	Salmonellosis		
S. typhi		Typhoid fever		
Shigella	0.1-1,000	Shigellosis		
Vibrio cholera		Cholera		
Vibrio spp. exc. V. cholera	10-10,000	Gastroenteritis		
Yersinia		Yersinosis		
Viruses				
Adenovirus	10-10,000	Respiratory disease, gastroenteritis, pneumonia		
Astrovirus		Gastroenteritis		
Norovirus (including Norwalk- like viruses)		Gastroenteritis		
Echovirus		Hepatitis, respiratory infection, aseptic meningitis		
Enterovirus (includes polio, enceph- alitis, conjunctivitis, and coxsackie viruses)	0.05-1000,000	Gastroenteritis, heart anomalies aseptic meningitis, polio		
Reovirus	0.1-125	Gastroenteritis		
Rotavirus	0.1-85,000	Gastroenteritis		
Parasites				
Cryptosporidium	3-13,700	Cryptosporidiosis		
Entamoeba	4-52	Amoebiasis (amoebic dysentery)		
Giardia	2-200,000	Giardiasis		

Source: US EPA (2004).

measured by a number of potential fecal indicator bacteria. There were no excess health effects observed at beaches with good quality water.

The microbial indicator group that showed the best relationship to swimming-associated gastrointestinal disease was the enterococci bacteria. These studies indicated that the illness rate among swimmers increased as the enterococci density increased. The studies also showed very clearly that illness in swimmers was associated with human or animal excreta. Although the pathogens causing the illness in swimmers were not identified, the symptoms-vomiting, nausea, and diarrhea-and illness duration for one to two days were very similar to those resulting from infection with enteric viruses.

Another epidemiological study that examined the relationship between contaminated coastal water and illness in swimmers was conducted in southern California (Haile et al., 1999). This study looked at the effect of runoff from storm drains that emptied onto coastal beaches. The highest densities of indicator bacteria were observed at the mouth of the drain, with levels decreasing with increasing distance from the source. Individuals of all ages who immersed their heads into water were potential subjects for this study. Nine to fourteen days following the beach interview, a follow-up

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Results of this study indicated that there were higher risks for gastrointestinal and respiratory symptoms when swimmers were closer to the storm drains, in water with a high density of indicator bacteria or a low ratio of total coliforms to fecal coliforms, and in water where enteric viruses were detected. The symptomatic end-points in this study were the same as those defined in previous US EPA studies and, as in these US EPA studies, the etiological agents responsible for illness and infections were not known. This study and previous US EPA studies clearly showed that there was a strong association between the level of fecal contamination in the water and the rate of illness in swimmers. Table 2 indicates that above densities of 35 enterococci per 100 ml, the illness rates were approximately 24 HCGI illnesses per thousand swimmers from the US EPA study, and 20 or more HCGI illnesses per thousand swimmers from the California study. These findings show that the contamination of coastal waters may pose a significant risk to individuals exposed to these waters.

SHELLFISH

Individuals who eat uncooked molluscan shellfish are also at risk if the shellfish are harvested from waters contaminated by

wastewater. Early in the 20th century, the most prevalent disease associated with shellfish was typhoid fever caused by the bacterium Salmonella typhi (Table 3). However, in the 1930s, water chlorination was introduced, and in the late 1940s antibiotics were used to treat infectious diseases. These practices were instrumental in lowering the number of illnesses, especially those transmitted by the fecal-oral route. The effectiveness of both of these practices can be seen in Table 3. From 1980 to 1990, the number of shellfish-associated outbreaks of disease caused by Salmonella typhi had decreased to zero.

Outbreaks where the causative agent was unknown appeared to increase in the 1980s relative to the number of cases observed before 1980. Between 1980 and 1990, a number of outbreaks occurred

trater as measured by manador bacteria					
Study	Illness ¹	Exposure (Enterococci/100 mL)	N	Illness Rate (per 1000)	Attributable Risk (per 1000) ²
Cabelli, 1983	HCGI	0 (non-swimmers)	9,162	12.3	
		≤ 35	11,937	20.9	8.6
		35+ - 104	1,348	37.1	24.8
		> 104	2,995	36.4	24.1
Haile et al., 1999	HCGI 1	≤ 35	7,689	6.5	
		35+ - 104	1,863	26.8	20.3
		> 104	857	58.3	51.8

Table 2. Epidemiological Studies of Swimmers' Risk: Gastrointestinal Illness Attributable to Swimming in Sewage Contaminated Water as Measured by Indicator Bacteria

¹ "Highly Credible Gastrointestinal Illness" indicated by the occurrence within 10 days (9-14 days for Haile et al. [1999]) of one of the following sets of symptoms: (1) vomiting; (2) diarrhea accompanied by a fever or disabling enough to remain home, remain in bed or seek medical advice; or (3) stomachache or nausea accompanied by fever. Haile et al. (1999) also reports results for an alternative definition for highly credible gastrointestinal (HCGI), not shown here.

² Swimming-associated illnesses per 1000 = difference in rate between swimmers and non-swimmers, or, for Haile et al. (1999), difference in rate between swimmers and minimally exposed (\leq 35 enterococci per 100 mL) swimmers.

Table 3. Shellfish-Borne Disease Cases and Outbreaks by Microbial Agent and Shellfish Species

	1894–1979		1980–1990		
	Cases	Outbreaks ⁴	Cases	Outbreaks	
Microbial Agent				•	
Salmonella typhi	3336	79	0	0	
Unknown/various ¹	2543	73	5852	174	
Hepatitis A	1724	36	104	7	
Salmonella	122	2	8	1	
V. parahaemolyticus	36	2	95	7	
Shigella	35	2	76	2	
Cholera non-01 ²	34	4	108	10	
Hepatitis c	16	1	1	0	
V. vulnificus	11	1	146	7	
Staphylococcus	5	1	0	0	
B. cereus	4	1	0	0	
V. cholera	2	1	14	1	
E. coli	2	1	0	0	
Vibrio spp.	1	0	1	0	
Norwalk virus	0	0	175	3	
Snow Mtn. virus	0	0	71	2	
Campylobacter	0	0	27	1	
Plesiomonas	0	0	18	1	
V. hollisae	0	0	15	0	
V. mimicus	0	0	14	0	
V. fluvialis	0	0	8	1	
Aeromonas	0	0	7	1	
V. alginolyticus	0	0	1	0	
Total	7,871	204	6,741	218	
Shellfish Species					
Oyster	3875	67	1815	74	
Hard clam	2539	77	3276	113	
Unknown/various ³	1212	46	1506	17	
Mussel	191	10	18	4	
Clamc	39	2	96	2	
Soft clam	15	2	28	7	
Scallop	0	0	2	1	
Total	7,871	204	6,741	218	

¹ Agent not identified or multiple agents reported

² Non-O1 serotypes

³ Type/species not specified

⁴ Two or more instances of the disease from the same source Source: Rippey (1991) that were caused by Norwalk virus and Norwalk-like viruses (viruses that cause gastrointestinal infection) that had not been reported previously. These apparent increases, however, may have been due to better surveillance and reporting techniques rather than actual increases in the number of outbreaks caused by microbial pathogens. Most of the outbreaks were associated with the consumption of oysters and clams, two molluscan species that are frequently eaten raw.

STANDARDS AND COASTAL WATER MONITORING

There are two approaches to controlling the risk associated with the discharge of sewage into coastal waters. First, the hazard may be controlled (i.e., wastewaters may be treated to eliminate the microbial pathogens that may be present therein); or second, exposure to waters contaminated with fecal material potentially containing microbial pathogens may be controlled. Although many authorities have taken steps to prevent hazards through the use of disinfectants, more effective treatment processes, and the construction of long-distance outfalls (pipes that discharge treated sewage into the ocean) that carry waste further offshore, away from human activities; the current approach in many jurisdictions is to control human exposure to wastewaters.

In the United States, standards for the quality of surface waters have been used since the first quarter of the 20th century. All of the standards have been based on the presence of feces in the water. Feces traditionally have been measured through the use of indicator bacteria, such as coliforms, fecal coliforms, *E. coli* and enterococci, which are found in very

high and constant densities in feces. The high densities of these organisms in feces allow them to be used as indirect measures of the amount of feces present in contaminated surface waters. Currently, enterococci are recommended for measuring the quality of marine waters in the United States.

As previously discussed, these microorganisms, which are enumerated in water samples using membrane filter procedures, have been shown to be correlated with gastrointestinal illness in swimmers (i.e., as the density of enterococci increase in the water, the gastrointestinal illness rate increases in swimmers). This relationship has been used to develop guidelines and standards for recreational water. Table 4 shows the US EPA recommended limits for marine waters in the United States. A geometric mean limit and a single sample limit are given for enterococci.

Most coastal states and municipalities in the United States routinely monitor recreational waters. The annual data collected from beach waters are regularly made available to the public. Table 5 lists the beach closings and postings of marine and estuarine beaches for the year 2004 in the United States. This shows that about 3.7 percent of the total estimated beach days were closed or posted in 2003 and that about 89 percent of the closings were due to a non-compliance with microbial limits. The other 11 percent of closings and postings were probably associated with preemptive actions associated with expected rainfall.

The microbial indicator used to measure the quality of shellfish-harvesting waters is fecal coliforms. Fecal coliforms are a heterogeneous group of bacteria that have common characteristics, such as a rod-shaped form, the ability to ferment lactose, and the ability to grow at a temperature of 44.5°C, which allow these organisms to be detected and measured. The standards for shellfish-harvesting waters have been in place since the mid-1930s. Early standards were based on the growth of coliforms in a series of 10 tubes containing 1 ml each of sample water. If growth occurred in more than half of the tubes, the limit would be

Table 4. Microbial Standards for Marine Water Quality Having Designated Uses of Recreation and Shellfish Harvesting

Use	Indicator	Classification	Limit	Description		
Geometric Mean Limits						
Recreation	Enterococci (CFU per 100 ml)	All Beaches	35	From at least 5 samples over a 30-day period		
Shellfish Harvesting	Fecal coliforms (MPN or CFU per 100 ml)	Approved ¹ / Conditionally Approved ²	14	At minimum from the most		
		Restricted ³ / Conditionally Restricted ²	88	(at least 2 or 5 samples per year ⁴)		
Single Samp	Single Sample/Percentile Limits					
2	Enterococci (CFU per 100 ml)	Designated Beach	104			
Recreation		Moderate FBC⁵	158	Single sample		
		Light Use FBC	276	maximum values		
		Infrequent Use FBC	501	CANE CONT		
Shellfish Harvesting	Fecal coliforms (MPN or CFU per 100 ml)	Approved/ Conditionally Approved	43 ⁶	10% of samples ⁸ or calculated 90 th		
		Restricted/ Conditionally Restricted	2607	percentile ⁹ not to exceed this limit		

¹ Approved shellfish growing area.

- ² Closed to harvesting based on a pollution event (rainfall, river flow, etc.).
- ³ By special license; shellstock subject to relaying or depuration after harvest.
- ⁴ 2 per yr. in remote status areas, 5 per yr. in areas impacted by point/non-point sources of pollution.
 ⁵ Full body contact.
- ⁶ MPN (most probable number) per 100 ml based on 5-tube decimal dilution; other limits apply for 3-tube decimal dilution (49), 12-tube single dilution (28), and membrane filtration (31 colony forming units [CFU]).
- ⁷ MPN per 100 ml based on 5-tube decimal dilution; other limits apply for 3-tube decimal dilution (300) and 12-tube single dilution (173).
- ⁸ When not impacted by non-point pollution sources.

⁹ If impacted by non-point pollution sources.

Sources: US EPA (1986) and National Shellfish Sanitation Program (2003).

	Coastal Beaches Monitored ¹			Closings & Advisories ¹		
	Number	Beach Days ²	Number	Beach Days	% of Total Beach Days	% Due to Bacteria ³
Alabama	26	3146	19	24	0.8	100
California	272	49776	992	4691	9.4	91
Connecticut	104	9464	96	178	1.9	11
Delaware	19	2052	1	5	0.2	100
Florida	303	110595	972	4816	4.4	100
Georgia	29	10585	77	412	3.9	100
Louisiana	15	2745	7	153	5.6	100
Maine	41	2501	15	56	2.2	100
Maryland	71	6532	29	197	3.0	90
Massachusetts	784	76832	243	738	1.0	100
Mississippi	21	7665	1	17	0.2	100
New Hampshire	15	915	3	6	0.7	100
New Jersey	228	28044	77	168	0.6	63
New York	310	33790	375	1108	3.3	30
N. Carolina	245	44100	47	363	0.8	100
Oregon	52	4732	16	432	9.1	100
Rhode Island	68	6188	28	766	12.4	100
S. Carolina	23	3519	130	395	11.2	24
Texas	49	7448	378	834	11.2	100
Virginia	49	5782	36	261	4.5	100
Washington	71	7597	9	72	0.9	71
Total	2,795	424,008	3,551	15,692	3.7%	89%

Table 5. Closings and Advisories for Marine and Estuarine Beaches Among Coastal States, 2004

¹ Excludes "permanent closures" (beaches closed 13 or more weeks).

² Number of beaches monitored x length, in days, of swimming season.

³ Percent of beach days closed or under advisory that were due to indicator bacteria levels in exceedence of standards.

Source: Natural Resources Defense Council (2005).

exceeded. Later on, mathematicians using probability theory determined that a water sample containing 70 coliforms per 100 ml would result in an average of half of the 10 tubes containing 1 ml of sample being positive. In the early 1970s, it was determined that 14 fecal coliforms per 100 ml was equivalent to 70 total coliforms per 100 ml. The resulting standard used by most jurisdictions today is that the geometric mean from at least 15 samples should not exceed 14 fecal coliforms per 100 ml and that no more than 10 percent of the time should a single sample exceed 43 fecal coliforms per 100 ml (see Table 4).

THE ECONOMICS OF SWIMMING-ASSOCIATED ILLNESS

The economic impact of illnesses associated with exposure to fecal-contaminated marine bathing waters was recently studied by Dwight et al. (2005). Their approach was to look at beach-going populations at two California beaches and to apply illness rates observed in independently conducted epidemiological studies to the California populations. Income data were obtained from local income surveys and medical expense data from a study by Nichol (2001). Only the swimmers whose illnesses were severe enough to cause them to lose work were considered. Total cost for four types of illnesses or infections observed in epidemiological studies was estimated for the California populations, including gastroenteritis, acute respiratory illness, ear infections, and eye infections. An estimated population at risk of approximately 5 million people, evaluated over a 31-month period, was used as a base for calculating the cost of illness for each illness/infection category.

The cost per illness by category, using 2001 dollars, was estimated to be US \$36.58 for gastroenteritis, US \$76.76 for acute respiratory disease, US \$37.86 for ear infections, and US \$27.31 for eye infections. Based on an annual estimate of 38,000 illnesses at the two beaches, the total cost for the four types of illness and infections would be about US \$3.4 million per year. The estimate for gastrointestinal illness alone was about US \$1.4 million per year.

These estimates, which the authors considered conservative, were based on illness rate data produced by Fleisher et al. (1998) in epidemiologic studies conducted in the United Kingdom. Fleisher et al. (1998) randomly assigned beachgoers to be immersed in the water or to sit on the beach without water contact with intensive monitoring of microbial indicators during the water exposure; they found an increased risk of gastrointestinal and other diseases at similar levels of enterococcus as the earlier US EPA studies. They also calculated what the health cost would be if the beaches met the US EPA and California standards for indicator bacteria for one year. Under these conditions, an expected 78,000 cases of swimming-associated gastrointestinal illness would occur. The public-health cost of these gastrointestinal illnesses would be about US \$2.87 million per year, and the total cost of all illnesses would be in excess of US \$7 million annually.

The authors correctly point out that only two health-cost variables, lost income and medical costs, were used to calculate the beach-exposure-related costs. Other costs related to self-medication or out-of-pocket costs for prescription medication might increase the total overall cost because most swimmers with gastroenteritis did not lose work days. This study provides evidence that the disease burden and the cost of that burden are probably quite high in the United States.

THE FUTURE OF MONITORING TO PROTECT PUBLIC HEALTH

The Beaches Environmental Assessment and Coastal Health Act of 2000 (P.L. 10-284, October 10, 2000) (Beach Act) directed states to begin monitoring beach waters using methods that were recommended by the US EPA in 1987 (US Congress, 2000). The method recommended for measuring marine beach water quality was method 1106.1 or method 1600 for enumerating enterococci using membrane filter procedures (US EPA, 2000). The Beach Act stimulated a renewed interest in the monitoring of marine bathing-beach waters, especially the interpretation of the data obtained through monitoring. This renewed interest occurred at a time when many states and municipalities were measuring water quality on a daily basis in an effort to better protect public health by providing more timely information to beach goers.

Normally, a water sample obtained from a beach is transported to the laboratory where the sample is filtered through a membrane that is then placed on a selective medium and incubated for twenty-four hours. After the incubation period, specific colonies are counted to determine water quality. Beaches are commonly closed or posted if the colony count exceeds the regulatory limit, even though the tests represent water quality of the previous day. A study conducted at beaches in southern California showed that the regulatory action taken on the day following the sample collection was incorrect about 70 percent of the time (Leecaster and Weisberg, 2001). Good public-health practice is not well served by error rates of this magnitude and this has stimulated a search for new ways of monitoring recreational waters. Some potential solutions for authorities that desire to protect public health on a daily basis are not available currently but may be in the near future.

One solution has been proposed by the World Health Organization (WHO) in a report popularly called the Annapolis Protocol (WHO, 1999). The Protocol suggests that all sources of fecal contamination that affect a beach resource be identified using sanitary surveys. If there are no fecal sources, there would be no need to monitor the water, except perhaps occasionally, or if the source or sources of fecal wastes cannot be remedied, the beach should not be used for swimming purposes until the source or sources are eliminated. For those beaches between these two extremes, a monitoring scheme can be used to determine the occurrence of any fecal events that may pose a risk to swimmers. This approach minimizes monitoring while protecting public health.

A second solution is to minimize the lag time so that the public can be notified in a timely manner whether the water is fit for swimming. Earlier warning can be accomplished by the use of newly developed, rapid quantitative polymerase chain reaction (QPCR) methods used to amplify DNA. The amplification of unique DNA target sequences from indicator bacteria to detectable levels can be accomplished in less than two hours and the results compared to a standard curve in order to quantify the results in terms of cell equivalents (Haugland et al., 2005). The US EPA is evaluating the QPCR approach to determine if this rapid method can be related to swimming-associated health effects (Wade et al., 2006).

A third solution would be to use a forecasting system based on meteorological or hydrographical data. Such data could be used to forecast water quality on a site-by-site basis at least one day into the future. Recent studies that addressed forecasting of water quality have showed promise that this approach may work, at least at certain locations (Ackerman and Weisberg, 2003; Olyphant and Whitman, 2004). These new approaches represent significant changes from the traditional monitoring schemes that have been practiced in the last century and promise new and better ways of controlling the exposure of individuals to recreational waters contaminated by human and animal excreta.

REFERENCES

- Ackerman, D. and S.B. Weisberg. 2003. Relationship between rainfall and beach bacterial concentrations on Santa Monica Bay beaches. *Journal of Water Health* 1(2):85–89.
- Cabelli, V.J. 1983. *Health Effects Criteria for Marine Recreational Waters*. EPA-600/1-80-031. US Environmental Protection Agency, Research Triangle Park, NC.
- Craun, G.F., R.L. Calderon, and M.F. Craun. 2005. Outbreaks associated with recreational water in the United States. *International Journal of Envi ronmental Health Research* 15(4):243–262.
- Dorfman, M. 2004. *Swimming in Sewage*. Natural Resources Defense Council, Environmental Integrity Project, New York, NY.
- Dwight, R.H., L.M. Fernandez, D.B. Baker, J.C. Semenza, and B. Olsen. 2005. Estimating the economic burden from illnesses associated with recreational coastal water pollution—A case study in Orange County, California. *Journal of Environmental Management* 76:95–103.
- Fleisher, J., D. Kay, M.D. Wyer, and A.F. Godfree. 1998. Estimates of the severity of illnesses associated with bathing in marine recreational waters contaminated with domestic sewage. *International Journal of Epidemiology* 27:722–726.
- Haile, R.W., J.S. Witte, M. Gold, R. Cressey, C.
 McGee, R.C. Millikan, A. Glasser, N. Harawa,
 C. Ervin, P. Harmon, J. Harper, J. Dermand, J.
 Alamillo, K. Barrett, M. Nides, and G.-Y. Wang.
 1999. The health effects of swimming in ocean water contaminated by storm drain runoff. *Epidemiology* 10(4):355–363.
- Haugland, R.A., S.C. Siefring, L.J. Wymer, K.P. Brenner, and A.P. Dufour. 2005. Comparison of *Entercoccus* measurements in freshwater at two recreational beaches by quantitative polymerase reaction and membrane filter culture analysis. *Water Research* 39:559–568.
- Leecaster, M., and S.B. Weisberg. 2001. Effect of sampling frequency on shoreline microbiology assessments. *Marine Pollution Bulletin* 42(11):1,150–1,154.
- Leeworthy, V.R., and P.C. Wiley. 2001. National Survey on Recreation and the Environment 2000. US Department of Commerce, National Oceanic and Atmospheric Administration, National Ocean Service, Silver Spring, MD.

- Natural Resources Defense Council. 2005. *Testing the Waters 2005: A Guide to Water Quality at Vacation Beaches.* Natural Resources Defense Council, Washington, D.C.
- National Shellfish Sanitation Program. 2003. *Guide* to the Control of Molluscan Shellfish. Interstate Shellfish Sanitation Conference. US Food and Drug Administration, Washington, D.C.
- Nichol, K. 2001. Cost benefit analysis of a strategy to vaccinate healthy working adults against influenza. *Archives of Internal Medicine* 161(5):749–759.
- Olyphant, G.A., and R.L. Whitman. 2004. Elements of a predictive model for determining beach closures on a real time basis: The case of 63rd Street beach, Chicago. *Environmental Monitoring Assessment* 98:175–190.
- Rippey, S.R. 1991. Shellfish Sanitation Program Technical Report: Shellfish Borne Disease Outbreaks. US Food and Drug Administration, Department of Health and Human Services, Washington, D.C.
- U.S. Environmental Protection Agency. 1986. Ambient Water Quality Criteria for Bacteria—1986.
 EPA 440/5-84-002. U.S. Environmental Protection Agency, Washington, D.C.
- U.S. Environmental Protection Agency. 2000. Improved Enumeration Methods for the Recreational Water Quality Indicators: Enterococci and Escherichia coli. EPA/821/R-97/004. U.S. Environmental Protection Agency, Washington, D.C.
- U.S. Environmental Protection Agency. 2004. *Report to Congress—Impacts and Control of CSOs and SSOs.* EPA 833-R-04-001. U.S. Environmental Protection Agency, Washington, D.C.
- Wade, T.J., R.L. Calderon, E. Sams, M. Beach, K.P. Brenner, A.H. Williams, and A.P. Dufour. 2006. Rapidly measured indicators of recreational water quality are predictive of swimming-associated gastrointestinal illness. *Environmental Health Perspectives* 114(1):24–28.
- World Health Organization (WHO). 1999. *Health-Based Monitoring of Recreational Waters: The Feasibility of a New Approach*, Geneva, Switzerland.