Risk Assessment: Exposure to Pathogens in Sewage-Contaminated Beach Sand

CAMRA 2006 QMRA Summer Insitute
(D. L. Greenberg, R. Ives, R. Jones, J. M. Proctor, T. Shibata, P. Gurian)

In mid-winter in a temperate area, untreated municipal sewage overflowed onto ocean beaches. To assess the infection risk posed to beachgoers by sewage-borne pathogens, we developed a framework to predict the concentration of pathogens in beach sand, and coupled the environmental concentration to sand-exposure parameters and dose-response relationships for representative sewage-borne pathogens. Our risk assessment focused on risk from the ingestion of contaminated sand, but other possible routes of pathogen exposure include ingestion of ocean water, inhalation of ocean spray, inhalation from wind-driven dispersal, consumption of shellfish, and penetration through dermal abrasions.

In our case scenario, raw sewage overflowed onto a public beach and approximately 238,000 L entered the ocean before a sand berm was constructed to impound the remainder. Approximately 3,230,000 L were recovered from the impound area and returned to the sewage treatment plant, while an estimated 2,270,000 L infiltrated an 8770 m^2 area of the beach. Sand samples were collected by the municipality and area public health organizations and analyzed for fecal coliform bacteria. After several weeks, the beach was eventually decontaminated with a dilute chlorine solution.

In our study, we model, evaluate, and compare the risk of pathogen exposure to the beach sand when (1) left untreated and (2) treated with chlorine. To validate the level of beach contamination predicted by our model, we compare the predicted fecal coliform bacteria concentration to the results of the municipality's sample analysis.

We used a traditional approach to risk assessment: hazard identification, exposure assessment, dose-response assessment, and risk characterization.

Hazard Identification

To determine the nature of microbial hazard, we examined data for the initial concentration of pathogens expected in sewage. Due to the numerous pathogenic and nonpathogenic microbes in sewage, and the lack of direct measurements, we chose fecal coliform bacteria as a general indicator, in order to compare estimates with the municipality's measurements, and four representative pathogenic organisms to focus our risk assessment: *Escherichia coli* O157:H7, *Shigella* species, *Cryptosporidium parvum*, and rotavirus.

Fecal coliform bacteria are commonly used in setting water standards, so *E. coli*, as a predominant member of this group, was selected to model risk of bacterial exposure, even though the four classes of recognized enterovirulent *E.coli* comprise a relatively small proportion of the 73,000 cases and 2,100 hospitalizations estimated to occur annually in the United States (CDC 1999).

Shigella species are close relatives of *E. coli*, and were selected for their high infectivity, though they are less prevalent than *E. coli* (CGER 1998).

Rotavirus is an enteric virus found in untreated municipal wastewater (CGER 1998) and is the most common cause of diarrheal disease in children worldwide (Current *et al.* 1996). The U.S. CDC estimates that rotavirus results in the hospitalization of approximately 55,000 children annually in the U.S., and immunity after infection is incomplete.

Cryptosporidium parvum is one of the most important enteric parasites present in the water environment. *Cryptosporidium* species infect the intestinal tract to cause diarrheal disease, and have a low infective dose. In immunocompetent individuals, cryptosporidiosis is usually self limiting, but for the immunocompromised, chronic cryptosporidiosis can become a serious and life-threatening disease (Keusch *et al.* 1995, APHA 1995). In a study of healthy human volunteers, the ID_{50} in healthy human volunteers of *C. parvum* 132 ingested oocysts (DuPont *et al.* 1995), but may be as low as 9 ingested oocysts (Okhuysen *et al.* 1999). These studies indicate that a low level of viable *Cryptosporidium* in the environment presents a health risk. Oocysts are the pathogen's environmentally transmissible stage and are infectious upon excretion (Korich *et al.* 1990).

Our risk assessment focused on ingestion of contaminated sand. Risk due to ingestion of contaminated water was not evaluated, as standard methods for monitoring are available and recreational water quality standards exist. The risk of inhalation of aerosolized pathogens was not assessed. There are pathogens in sewage, such as *Legionella*, that we might expect to be an inhalation hazard, but no appropriate sampling for pathogenic aerosols was conducted in our case scenario and the exposure prediction is complex. To further limit the scope of the risk analysis, exposure to pathogens via consumption of shellfish and infection of dermal abrasion were not evaluated.

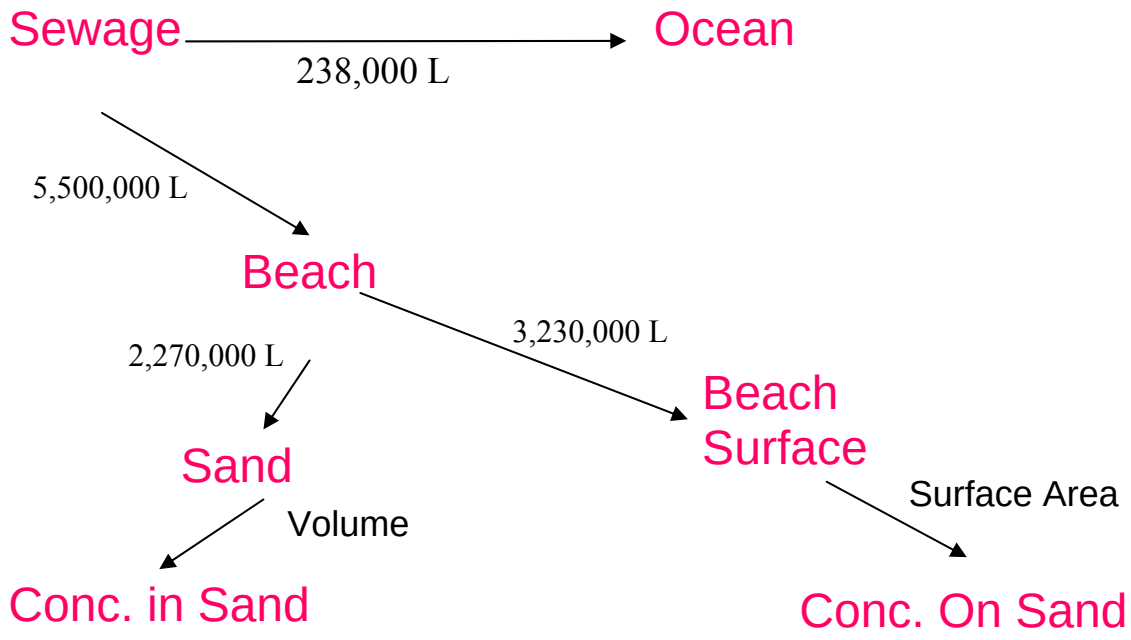

Figure 1. Framework for the environmental dispersion of raw sewage spilled onto an ocean beach.

Exposure Assessment

To assess the infection risk posed to beachgoers by sewage-borne pathogens, we developed a framework (Figure 1) to predict the concentration of pathogens in beach sand, and considered exposure to contaminated sand. The municipality estimated that at one of the six contaminated sites, 238,000 L of sewage directly entered the ocean. Of the 5,500,000 L remaining at the beach, about 3,230,000 L were estimated to have been removed from the sand surface during the vacuuming of the debris/scum, and the remaining 2,270,000 L infiltrated the sand. We assumed that the microbes were uniformly distributed within the sewage, such that the proportions of pathogens in the three environmental media (ocean, bermed pond, and infiltration) are proportional to the respective sewage volumes. The spill at this site covered 8770 m^2. Environmental measurements of fecal coliform bacteria were made at depths of 1 ft and 3 ft. Therefore, we assumed that the sewage-borne microbes could be well mixed in the sand to a depth of 30 cm and 100 cm. To enable health-conservative risk estimates, we also predicted the pathogen concentration in sand when pathogens were well-mixed to a depth of 1 cm.

We expect that most microbes are associated with the sediment component of sewage due to the pH of the sewage and charges of the microbes and sediment; and that more of the sediment component of the sewage would remain on the surface of the beach rather than penetrate the sand due to the relative sizes of the sediment particles and sand pores. Therefore, we expect our assumption of uniform distribution overestimate the number of pathogens which infiltrate the beach sand.

Table 1. Parameters for Exposure and Dose-Response Assessment

Organism	Conc. In Sewage (CFU/100ml)	Atten. Rate (log_{10}/d)	Cl_2 Attn. Rate (log_{10}/d)	Dose-Response Relationship
Fecal coliform group	10^2-10^5 [A]	-0.0088 [B]	-9000 [C]	~ Beta Poisson (α = 0.1778, N_{50} = 8.6E7) [D]
E. Coli O157:H7	10^2-10^4 [E]	-0.0088 [B]	-9000 [C]	~ Beta Poisson (α = 0.49, N_{50} = 5.96E5) [F]
Shigella	100-10^3 [A]	-0.0066 [G]	-240 [H]	~ Beta Poisson (α = 0.2100, N_{50} = 1120) [D]
C. parvum	10^2-10^5 [A]	-0.0225 [I]	0 [J]	~ Exponential (λ = 0.004) [D]
Rotavirus	10^3-10^4 [A]	-0.036 [K]	-900 [L]	~BP (α=0.26, β=0.42) [D]

[A] (CGER 1998)
[B] (Craig, Fallowfield, & Cromar 2002), fecal coliform assumed to be representative of *E. coli* O157:H7
[C] (Rice, Clark, & Johnson 1999), *E. coli* assumed to be generalizable to all fecal coliform bacteria
[D] (Haas, Rose, & Gerba 1999)
[E] (Haas 2006, unpublished communication)
[F] (Haas *et al.* 2002)
[G] *Shigella sonnei* (John 2003), linearly interpolated for temperature
[H] (King *et al.* 1988)
[I] (Nasser, Tweto, & Nitzan 2006)
[J] (Lisle & Rose 1995)
[K] (Azadpour-Keeley & Ward 2005), by analogy with polio virus for sand
[L] (Vaughn, Chen, & Thomas 1986)

Typical concentrations of microbes in raw sewage are indicated in the second column of Table 1. We assumed that the concentrations had log uniform distribution within the range indicated in Table 1.

To account for the loss of viability in the environment, we incorporated into our model empirical attenuation rates for fecal coliform and our representative pathogens (Table 1). In some cases, we were unable to locate published rates for ocean sand, so rates were selected to best fit the soil type, temperature, and pH conditions, with favor given to slower attenuation rates that would provide health-conservative risk assessments. Rates were also collected for the representative organisms in the presence of free chlorine (Table 1) to model a scenario with chlorine treatment. The municipality used chlorine as their effective treatment to disinfect sewer overflow onto the beach as recommended by EPA guidelines (EPA 1999), and we wanted to predict risk associated with this solution.

Our model could be used to conduct simulations for several different treatment methods such as ozone, chloride dioxide, or aeration, but we ruled these out. The gases, while very effective at disinfection, present difficulties in the logistics of their use in the beach scenario due to the large surface area that would be needed to be covered, an inadequate understanding of gas penetration into the subsurface of sand, and a high cost associated with these methods. In addition, small experiments conducted on a test plot by the municipality indicated that aeration was ineffective at reducing pathogen numbers. In contrast, the chlorination solution provides a cheap, fast, and effective measure to cover the largest area with penetration to greatest depth.

We focused on the ingestion route of exposure. Children comprise one of the most susceptible groups to infection, and they are the most likely to be playing in the sand of a beach (WHO 2003). Ingestion of soil is a major potential route of human exposure to hazards on the contaminated beach, especially children as a result of childhood behavior (EPA 1997). Children at contaminated beaches are at great risk from the hazards, especially children younger than 7 years old, who are most likely to ingest soils by hand-to-month behaviors (Freeman *et al.* 2001). Since children aged from 0 to 1 year old are presumed to have limited activities (Zartarian *et al.* 2005), children from 2 to 6 years old are the most susceptible population in our model.

Quantifying the amounts of sands children may ingest during time they spend at the beach would vary based on many environmental and other conditions, such as temperature, humidity, water content in the sand, hand sizes, and hand-to-mouth behaviors. The amounts of microbes that children may intake can be estimated based on the estimated amounts of sand ingestion. The estimated amounts of microbes to which children may be exposed through oral ingestion of contaminated soil can be calculated based on the following equation.

$$M_{ingestion} = M_{soil} \cdot SS \cdot H \cdot HtoM \cdot T$$

In this equation, $M_{ingestion}$ is an estimated amount of microbe that a child could ingest; M_{soil} is a concentration of the microbe in a contaminated soil (CFU/g of soil); SS is the soil-skin adherence factor (mg/cm^2), a required parameter for calculating dermal dose when the exposure scenario involves dermal contact with hazards in soil (EPA 2002); H is the area of a hand (cm^2); $HtoM$ is a frequency of hand-to-mouth activity (hr^{-1}); and T is

a time of exposure at the site (hr). For the *SS*, we assume a geometric mean of 0.14 (Zartarian *et al.* 2005), originally used for risk assessment on children at a playground. However, the *SS* of 0.14 may be not valid of the sand is wet or contains organics because it will increase the adherence of soil to the hands. For the *HtoM*, documentations of frequency of hand-to-mouth behavior are very limited, but a geometric mean of 8.5, which was also used for the playground study (Zartarian *et al.* 2005), is assumed to be reasonably accurate. It must be noted that all soil may not be ingested by the hand-to-mouth behavior. On the other hand, it is conservative to assume that all microbes in the soils on the hands are ingested through the childhood behavior. For the *H*, data on surface area of the hands of children (aged 2 to 6 years old) are also limited. An estimation of *H* can be calculated based on age, height, and weight. For the *T*, the length of time specific age groups of children spend at beach has not been well documented yet. Overall, sufficient values of those parameters for the equation could not be obtained for this case study.

Therefore, the amounts of microbes that children may intake at the contaminated beach were calculated based on a daily soil ingestion rate for children. The U.S. EPA recommended use of 100 mg/day for risk assessment on children associated with soil ingestion (EPA 1997). To increase the health-protection in our risk assessment, we described the ingestion of contaminated sand by a log-normal distribution with a geometric mean of 200 mg/day and geometric standard deviation of 3. This distribution does not account for extreme pica behavior, for which the U.S. EPA assumes a daily ingestion of 10,000 mg/day of soil.

Other parameters that we varied in the model were sand depth and time. We assumed in separate trials that the microbes were well mixed in the sand (uniformly over the entire spill area) to the depth of 1 cm, 30 cm, and 100 cm, and for each depth predicted the concentration of microbes per gram dry weight of sand from the time of the spill (day 0) to day 33. We expect most pathogens to be captured at or near the beach surface, especially virus particles, which because of their charge are attracted to soil particles at neutral or near-neutral pH (Hijnen *et al.* 2005). Wang *et al.* (1981) observed that viruses were retained by sand columns in the first 7 cm. Our worst-case scenario is that all organisms are concentrated within the first centimeter, thereby making them most accessible to hand-to-mouth behavior.

Table 2. Predicted and Observed Concentrations of Fecal Coliform Bacteria

Time (days)	Depth (cm)	Median Predicted Concentration (10^{th}, 90^{th} %) (CFU/gdw)	Measured Concentration Range
11	30	2800 (456, 18,000)	5100–11,000 MPN/gdw
	100	865 (264, 5400)	310,000 MPH/gdw
22	30	14 (2.2, 89)	1800–12,000 CFU/gww
	100	4 (0.67, 27)	200–2500 CFU/gww
33	30	0.07 (0.01, 0.44)	< 17 CFU/gww
	100	0.02 (0, 0.13)	< 2 CFU/gww

Time was considered in our models due to model the real-world clean-up process. We predicted the distribution of fecal coliform bacteria and the representative bacteria at time points ranging up to 33 days after the release, in order to compare the predicted concentrations of fecal coliform bacteria to the concentrations measured by the municipality. As indicated in Table 2, the fecal coliform bacteria concentration predicted by our model was similar to that measured on post-release day 11, with the variation easily accounted for by our use of log-uniform distribution for the initial concentration. Our model under-predicts the concentrations at later times, but our results below suggest that the critical period for health risks ends around day 11.

Dose-Response Relationship

Dose-response functions for our representative pathogens were identified from the literature, and are indicated in Table 1. Even though we encountered multiple (and often widely varying) sets of parameters for each pathogen in our literature search, we chose to keep the model for this study simple by not including a distribution of distributions.

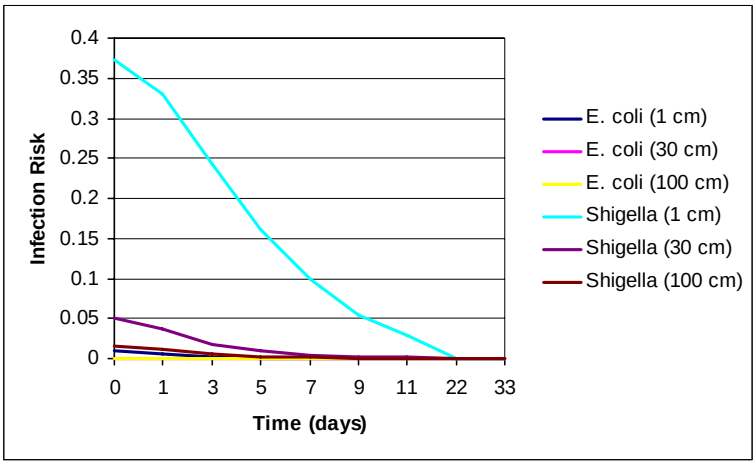

Figure 2. Predicted Bacterial Infection Risk

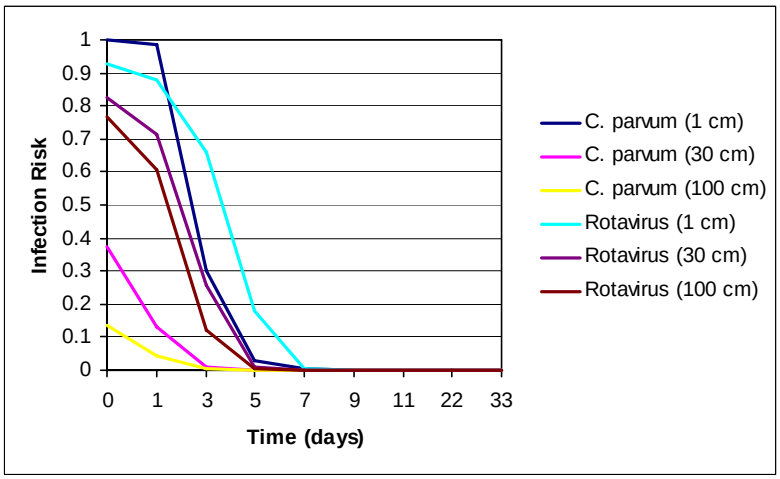

Figure 3. Predicted Non-bacterial Infection Risk

Risk Characterization

When the exposure model is integrated with the route of exposure and the dose-response relationships, the infection risk appears to be primarily from potential exposures to rotavirus and *C. parvum* during the first week (168 hours) post-release (Figure 2 and Figure 3), while the risks from *E. coli* and *Shigella* species are maximially 0.05 per day unless the *Shigella* are assumed only to penetrate the top 1 cm of the sand. Notably, for all representative pathogens and all scenarios (except the 1 cm depth for *Shigella*), the infection risk is very low.

Though the data are not shown here, our model predicts that for all representative pathogens except *C. parvum*, exposure to free chlorine results in the elimination of infection risk immediately. The primary obstacle to this method of treatment is the difficulty of making free chlorine available at depths of 1 m or more in sewage-saturated sand.

Discussion

There is no standard for the concentration of microbes in ocean beach sand, nor for the infection risk. The most similar standard we were able to identify was the marine bathing water standard of the U.S. EPA: 1.9% cumulative risk of illness per bather day. Our model predicts that *Shigella* in 1 cm of sand (our worst case) reaches this level with 90% confidence after 18 days. The other representative pathogens reach this level in less than 10 days. These predictions suggest that given the removal of the debris/scum layer and no further treatment, it will be possible to attain the marine water bathing standard in approximately three weeks. Our model, however, is limited by its reliance on pathogen-specific risks, rather than cumulative risks.

After the overflow event, the municipality elected to apply dilute chlorine on the ocean beaches contaminated with sewage. Our model suggests that chlorine treatment sufficient to produce free chlorine in the sand will rapidly reduce the concentrations of *E. coli*, *Shigella*, and rotavirus. Chlorine, however, does not adversely impact the viability of *C. parvum*. Therefore, the attenuation of *C. parvum* is the limiting factor in opening the beach subsequent to chlorine treatment. Our model predicts that in approximately 8 days, *C. parvum* risk is reduced to below the marine bathing water standard with 90% confidence. Therefore, given immediate chlorine treatment, the beach sand would attain the marine bathing water infection risk standard 8 days post-release.

Our assessment suggests that the municipality responded appropriately to the sewage overflow onto the ocean beaches. In particular, it was important that the municipality was able to remove the debris/scum layer rapidly. Had additional pathogens leached into the soil, clean-up efforts would have encountered far greater concentrations, at greater depths, than the real case. As evidenced by the municipality's monitoring data, free chlorine was generated to a depth of 3 ft and was able to reduce the concentration of fecal coliform bacteria in sand. Some monitoring data not shown here, however, indicate that the microbial concentration may rebound, though the increased concentrations may have resulted from heterogeneity in environmental conditions. As a result of this heterogeneity, we would recommend, for the sake of the protection of public health, that the closure of the beach be extended beyond the 8 or 18 days indicated by the options above so that environmental monitoring can demonstrate prolonged absence of sub-surface pathogenic microbes.

One alternative that might be explored through a risk assessment process is the flushing of sewage into the ocean, rather than entrapment within a berm. Construction of a berm could permit infiltration of pathogens into the sand in excess of that which would occur had the sewage not been pooled on the sand. Certainly, this approach would significantly increase the pathogen load into the ocean. If the local currents and tides are such that the contamination would be quickly returned to the beach, even over a larger area, this is clearly not advisable. Depending upon turbulence and flow at the particular site, however, the pathogens could be rapidly diluted.

Uncertainties and Limitations

The uncertainties and limitations in our exposure model and risk characterization can be grouped into three categories:

Availability of Source Data

Our model used attenuation rates for organisms measured in soils other than sand, for temperature, pH, and humidity conditions other than those that existed at the time of the spill. We encountered similar gaps in published data for the effects of chlorine and other treatments, and such a wide variety of dose-response models that it would be irresponsible (even with expert guidance) to claim that we are certain of the predicted risk even if the concentration models are correct. We feel that this lack of consistent data is potentially the greatest obstacle to reliable quantitative microbial risk assessment.

The Complexity of Our Model

We assumed, for simplicity, that the absorbed water infiltrated the sand evenly across the entire affected area. Further, we assumed that the microbes were distributed evenly in that water, and that the process of infiltration into the sand would mix them uniformly through the depths we tested. We recognize that the hydrology of a sewage plume on an ocean beach is far more complex than this, but we lacked the expertise and information about the physical and hydrological properties of the site to use any more reliable estimation technique.

Our model only addresses one of the seven routes of infection that we identified. We selected the one we believed would have the highest risk, but a thorough study should include the others to generate a cumulative model. Similarly, our model highlights four pathogenic organisms, when there are thousands present in sewage. Broad-spectrum attenuation, treatment response, and infectivity equations seem unlikely, if not impossible, but better techniques for estimating cumulative totals would clearly help researchers and practitioners evaluate potentially expensive treatment options.

Our dose-response equations do not necessarily generate realistic estimates for infectiousness in children, who may be more susceptible. Further, we greatly simplified hand-to-mouth behavior (for lack of data), and underestimated the possibility of pica behavior. Future work in toward determining a reliable dose-response model for contaminated beach sand is suggested.

Results, Evaluation, and Recommendations

Our initial results underestimated the concentration of fecal coliform bacteria,

especially at the deeper test depth and longer times after the overflow. Some of this may be accounted for by our assumption of log-uniform distribution on initial concentration, as well as the reduced complexity of our hydrologic model, but we believe there may be regrowth, ocean backwash, and external contamination factors that we did not consider.

The municipality's tests only for fecal coliform bacteria, at limited times and unknown locations within the affected area, severely limited our ability to evaluate and refine the pathogenic concentration models. While we recognize the high cost associated with greatly increased monitoring of disaster sites, we feel that the relevant governmental agencies should budget for that expense and work with centers such as CAMRA and local research institutions to conduct thorough studies of affected areas in order to understand the microbial fate and transport processes at work. Three tests for one indicator group over the space of a month do not provide enough data to significantly inform future clean-up efforts.

Finally, our recommendation to apply chlorine has its own environmental consequences that we did not include in the risk assessment. Most obvious, of course, is that a high concentration of residuals may be as toxic to the humans as the pathogens. In discussing the case, we questioned whether the municipality was correct in waiting for three weeks before conducting large-scale chlorine treatment, and reached the conclusion that it may have been their best way to ensure that the cure would not be worse than the disease. Ultimately, we decided that they were correct to have bermed the sewage, which enabled them to remove more than 50% of it with no adverse effect on the environment, but that they could have begun their chlorination regimen sooner. However, without filling all the data gaps above, it would be impossible to accurately model the effectiveness of a different approach.

References

American Public Health Association (APHA), American Water Works Association (AWWA), and Water Environment Federation (WEF). 1995. *Standard Methods for the Examination of Water and Waste Water* 19th Edition. Section 9711.

Azadpour-Keeley A and Ward CH. 2005. Transport and Survival of Viruses in the Subsurface—Processes, Experiments and Simulation Models. *Remediation* Summer 2005:131–138.

Centers for Disease Control and Prevention (CDC). 1999. Emerging Infectious Diseases. National Center for Infectious Diseases, Atlanta GA. Volume 4, Issue 4, October-December 1999.

CGER: The National Research Council Commission on Geosciences, Environment and Resources. 1998. *Issues in Potable Reuse: The Viability of Augmenting Drinking Water Supplies with Reclaimed Water*. National Academy Press, Washington DC.

Craig DL, Fallowfield HJ, Cromar NJ. 2002. Comparison of Decay Rates of Faecal Indicator Organisms in Recreational Coastal Waters and Sediment. *Water Science and Technology: Water Supply* 2(3):131–138.

Current WL. 1987. *Cryptosporidium*: Its Biology and Potential for Environmental Transmission. *CRC Critical Reviews in Environmental Control* 17:21-31.

DuPont HL, Chapell CL, Sterling CR, Okhuysen PO, Rose JB, Jakubowski W. 1995. The Infectivity of *Cryptospodium Parvum* in Healthy Volunteers. *New England Journal of Medicine* 332(13):855.

Environmental Protection Agency (EPA). 1997. *Exposure Factor Handbook*. U.S. EPA, National Center for Environmental Assessment, Office of Research and Development, Washington, D.C.

Environmental Protection Agency (EPA). 1999. Combined Sewer Overflow Technology Fact Sheet: Chlorine Disinfection. EPA Report 832-F-99-034.

Environmental Protection Agency (EPA). 2002. *Child-Specific Exposure Factor Handbook*. Interim report EPA-600-P-00-002B.

Freeman NCG, Jimenez M, Reed KJ, Gurunathan S, Edwards RD, Roy A, Adgate JL, Pellizzari ED, Quackenboss J, Sexton K, Lioy PJ. 2001. Quantitative Analysis of Children's Microactivity Patterns: The Minnesota Children's Pesticide Exposure Study. *Journal of Exposure Analysis and Environmental Epidemiology* 11(6):501–509.

Haas CN, Rose JB, Gerba CP. 1999. *Quantitative Microbial Risk Assessment*. John Wiley & Sons, New York NY.

Haas CN, Thayyar-Madabusi A, Rose JB, Gerba CP. 2000. Development of a dose-response relationship for *Escherichia coli* O157:H7. *International Journal of Food Microbiology* 56(2-3):153–159

Hijnen WAM, Brouwer-Hanzens AJ, Charles KJ, Medema GJ. 2005. Transport of MS2 phage, *Escherichia coli*, *Clostridium perfringins*, *Cryptosporidium parvum*, and *Giardia intestinalis* in a Gravel and a Sandy Soil. *Environmental Science & Technology* 39(20):7860-7868.

John, DE. 2003. Transport and Survival of Water Quality Indicator Microorganisms in the Ground Water Environment of Florida: Implications for Aquifer Storage and Waste Disposal. Doctoral dissertation, College of Marine Science, University of South Florida, Tampa FL.

Keusch GT, Hamer D, Joe A, Kelly M, Griffiths J, Ward H. 1995. Cryptosporidia—Who Is at Risk? *Schweizerische Medizinische Wochenschrift* 125(18):899–908.

King CH, Shotts EB Jr., Wooley RE, Porter KG. 1988. Survival of Coliforms and Bacterial Pathogens within Protozoa during Chlorination. *Applied and Environmental Microbiology* 54(12):3023–3033.

Korich DG, Mead JR, Madore MS, Sinclair NA, Sterling CR**.** 1990. Effects of Ozone, Chlorine Dioxide, Chlorine, and Monochloramine on *Cryptosporidium parvum* Oocyst Viability. *Applied and Environmental Microbiology* 56(5):1423–1428.

Lisle JT, Rose JB. 1995. *Cryptosporidium* Contamination of Water in the USA and UK: a mini review. Aqua 44(3):107-117 1995.

Nasser AM, Tweto E, Nitzan Y. 2006. Die-off of *Crytosporidium parvum* in Soil and Wastewater Effluents. Journal of Applied Microbiology.

Okhuysen PC, Chappell PL, Crabb JH, Sterling CR, DuPont HL. 1999. Virulence of Three Distinct *Cryptosporidium parvum* Isolates for Healthy Adults. *Journal of Infectious Disease* 180(4):1275–1281.

Rice EW, Clark RM, Johnson CH. 1999. Chlorine Inactivation of *Escherichia coli* O157:H7. *Emerging Infectious Diseases* 5(3):461–463.

Vaughn JM, Chen YS, Thomas MZ. 1986. Inactivation of Human and Simian Rotaviruses by Chlorine. *Applied and Environmental Microbiology* 51(2):391–394.

Wang DS, Gerba CP, Lance JC. 1981. Effects of Soil Permeability on Virus Removal through Soil Columns. *Applied and Environmental Microbiology* 42(1):83–88.

World Health Organization (WHO). 2003. *Guidelines for Safe Recreational Waters Vol. 1 – Coastal and Fresh Waters*. Chapter 6, p122.

Zartarian VG, Xue J, Özkaynak H, Dang W. 2005. *A probabilistic exposure assessment for children who contact CCA-treated playsets and decks: Using the stochastic human exposure and dose simulation model for the wood preservative exposure scenario (SHEDS-Wood), Final Report,* U.S. Environmental Protection Agency, Office of Research and Development, National Exposure Research Laboratory and Office of Pesticide Programs, Antimicrobials Division, Washington DC.