Appendix A

TYPES OF PATHOGENS & TYPES OF PATHOGEN INDICATOR BACTERIA

APPENDIX A
TYPES OF PATHOGENS & TYPES OF FECAL INDICATOR BACTERIA

A.1 TYPES OF BACTERIA

Pathogens most commonly identified and associated with waterborne diseases can be grouped into the three general categories: bacteria, protozoans, and viruses (U.S. EPA 2001).

A.1.1 BACTERIA

Bacteria are microscopic unicellular organisms ranging from approximately 0.2 to 10 micrometers (µm) in length. They are distributed ubiquitously in nature, including the intestinal tract of warm-blooded animals. Many types of harmless bacteria colonize the human intestinal tract and are routinely shed in feces. In addition, pathogenic (disease-causing) bacteria, such as verotoxigenic *E. coli* (including serotype 0157:H7), *Salmonella*, and *Campylobacter*, are present in the feces of infected humans and animals and can contaminate surface water and groundwater as a result of inadequate waste treatment or disposal methods. Many groups of intestinal bacteria, including the coliform and enterococci groups, have historically been used as an indication that an environment has been contaminated with human sewage.

A.1.2 PROTOZOANS

Protozoans are unicellular organisms that are present primarily in the aquatic environment. Of the 35,000 known species of protozoans, almost 30 percent are pathogenic. Pathogenic protozoans can occur in humans and animals where they multiply in the intestinal tract of the infected individual or animal and are later excreted in feces as cysts. Protozoan cysts do not reproduce in the environment, but are capable of surviving dormant in the soil and surface water for extended periods of time, which makes them a prominent public health concern.

Two waterborne protozoans of major public health concern are *Giardia lamblia* and *Cryptosporidium parvum*. The *Giardia* organism inhabits the digestive tract of a wide variety of domestic and wild animal species, as well as humans. Once shed in feces, G*iardia* cysts are frequently found in rivers and lakes. Infection by *Giardia* can result in giardiasis in humans, which is characterized by gastroenteritis, particularly among the young and elderly. *Giardia* is considered nonpathogenic in cattle because it is usually found in animals that have normal feces and no sign of disease. However, among the human population, giardiasis affects approximately 200 million people worldwide and is one of the most prevalent waterborne diseases in the United States. *Cryptosporidium* species are a group of parasitic protozoa that are recognized as pathogens of domesticated livestock, poultry, and wildlife and are readily transmitted to humans. *Cryptosporidium* oocysts are about 4-6 µm in diameter, slightly larger than bacteria, and relatively unaffected by conventional

methods of wastewater disinfection, such as chlorination. Infection by *Cryptosporidium* can cause cryptosporidiosis, whose symptoms include loss of appetite, nausea, and abdominal pain followed by acute or persistent diarrhea. Although *Cryptosporidium* infections are usually of short duration and self-limiting in individuals with an intact immune system, there is no specific treatment available and the infection can be life threatening in patients with profound impairment of immune function.

A.1.3 VIRUSES

Viruses are obligate intracellular parasites, incapable of replication outside of a host organism. They are very small, ranging from 0.02 to 0.2 µm. Viruses that are of a public health concern are viruses that replicate in the intestinal tract of humans, and are referred to as human enteric viruses. Sewage overflows and improperly functioning sewage systems are considered to be primarily responsible for water contamination. Individuals can become infected through consumption of contaminated water, swimming in contaminated water, or through person-to-person contact with an infected person. Symptoms of infection include vomiting and diarrhea, with the severity of disease and mortality increasing in older age groups. The most significant human enteric viruses include hepatitis A, rotaviruses, noroviruses, adenoviruses, enteroviruses, and reoviruses.

A.2 TYPES OF PATHOGEN INDICATOR BACTERIA

Several groups of intestinal bacteria have been used as indicators that a waterbody has been contaminated with human sewage and that pathogens are present. Most strains of pathogen indicator bacteria do not directly pose a health risk to swimmers and those recreating in the water, but indicator bacteria often co-occur with human pathogens and are easier to measure than the actual pathogens that may pose the risk of illness. It is impractical to directly measure the wide range of types of fecal-borne pathogens (bacteria, viruses, and protozoans) and the methods to detect human pathogens are characteristically expensive and inefficient, or may be not available. Indicator bacteria are described in Chapter 2 and include:

A.2.1 TOTAL COLIFORM BACTERIA

Total coliforms are a group of bacteria that are widespread in nature. All members of the total coliform group can occur in human feces, but some can also be present in animal manure, soil, submerged wood, and other places outside the human body. Thus, the usefulness of total coliforms as an indicator of fecal waste contamination depends on the extent to which the bacteria species found are fecal and human in origin. Because total coliforms can come from non-fecal sources, they are no longer recommended as an indicator for assessing the support of recreation beneficial uses (U.S. EPA 1986). However, total coliform is still recommended for use in assessing support of shellfish consumption based on criteria adopted in 1925. The shellfish criteria are based on investigations made

by the U.S. Public Health Service that assessed the occurrence of typhoid fever or other enteric diseases attributed to shellfish harvesting (U.S. FDA 2011).

A.2.2 FECAL COLIFORM BACTERIA

Fecal coliform bacteria are a subgroup of total coliform bacteria found in the intestinal tracts of animals, and thus, are considered a more specific indicator of fecal waste contamination of water than the total coliform group. Fecal coliform bacteria concentration criteria were initially recommended by U.S. EPA (1976) for assessing support of recreational use. However, since 1976, several key epidemiological studies were conducted to evaluate the criteria for effectiveness at protecting public health from water contact recreation (Cabelli et al. 1982; Cabelli et al. 1983; Dufour 1983; Favero 1985; Seyfried et al. 1985a, Seyfreid et al. 1985b) The studies concluded that the U.S. EPA (1976) recommended fecal coliform bacteria criteria had no scientific basis. As a result of the new information derived from epidemiological studies, the U.S. EPA (1986) changed the criteria recommendation to use the pathogen bacteria indicators of *E. coli* and enterococci bacteria, instead of fecal coliform bacteria.

In addition, detection of fecal coliform bacteria in recreational waters may overestimate the level of fecal waste contamination because this bacteria group contains several genera that are not of fecal origin (e.g., Enterobacter, Klebsiella, Citrobacter). For example, *Klebsiella* bacteria are commonly associated with soils and the surfaces of plants, so that areas with allochthonous organic debris may show high levels of fecal coliform bacteria that do not have a fecal-specific bacteria source.

A.2.3 *ESCHERICHIA COLI* (E. COLI) BACTERIA

E. coli is a species of fecal coliform bacteria that is specific to fecal material from humans and other warm-blooded animals. U.S. EPA (2012) compiled numerous epidemiological studies and concluded that *E. coli* bacteria another indicator of human health risk from water contact in recreational freshwaters. The criteria are established for both the geometric mean and the statistical threshold value (STV). The geometric mean criterion is compared to the logarithmic average of the bacteria concentration distribution. The STV criterion is compared to the 90th percentile of the bacteria concentration distribution.

Criteria were published for two different levels of illness risk (Table A.1). The first level of risk (36 estimated illnesses per 1,000 recreators) is the same risk level applied with the previous recreational criteria (i.e., USEPA 1986). The 1986 U.S. EPA criteria correspond to the level of risk associated with an estimated illness rate of the number of highly credible gastrointestinal illnesses (HCGI) per 1,000 primary contact recreators. The information developed for the 2012 U.S. EPA criteria use a more comprehensive definition of GI illness, referred to as NEEAR-GI (NGI), which includes diarrhea without the requirement of a fever. Because NGI is broader than HCGI, more illness cases were reported and associated with recreation using the NGI definition of illness, at the same level of water quality observed using the previous illness definition (i.e., HCGI). The U.S. EPA (2012) also recommends

criteria that correspond to an illness rate of 32 NGI per 1,000 primary contact recreators to "encourage an incremental improvement in water quality."

The 2012 U.S. EPA criteria are expressed as colony-forming units per sample volume (cfu/100mL) based on membrane filtration methods (USEPA 2002a; USEPA 2002b). Many laboratories, including the Regional Water Board Microbiology Laboratory, use a different analysis method to measure *E. coli* (and *Enterococcus*) bacteria concentrations (IDEXX 2001). These methods, (Colilert® and Enterolert® Quanti-Tray/2000) have been shown to produce equivalent results as the membrane filtration methods (Budnick et al. 1996; Yakub et al. 2002) and have been approved by the USEPA in the Code of Federal Regulations (40 CFR 136.3).

Table A.1. Recreational Water Quality Criteria for *E. coli* Bacteria Concentrations (USEPA 2012)

Pathogen Indicator Bacteria	**Recommendation 1** Estimated Illness Rate 36 per 1,000 recreators		**Recommendation 2** Estimated Illness Rate 32 per 1,000 recreators	
	Geometric Mean (cfu/100mL)	Statistical Threshold Value (cfu/100mL)	Geometric Mean (cfu/100mL)	Statistical Threshold Value (cfu/100mL)
E. coli	126	410	100	320

A.2.4 ENTEROCOCCI BACTERIA

Enterococci bacteria are a subgroup within the fecal streptococcus bacteria group. enterococci bacteria are distinguished by their ability to survive in salt water, and therefore more closely mimic pathogens than the other indicators in marine environments. U.S. EPA (2012) recommends enterococci bacteria concentration as another bacteria indicator of human health risk in salt water for recreation.

U.S. EPA (2012) states that enterococci bacteria concentrations may also be used as an indicator of human health risk in fresh water. Similar to *E. coli* bacteria, the enterococci bacteria criteria are established for both the geometric mean and the STV for protection of water contact recreation (Table A.2). The criteria are based on epidemiological studies at U.S. beaches. The studies enrolled participants at a number of beach study sites and followed them to compare incidence of illness between the exposed (swimmers) and unexposed groups. Exposed groups involved swimmers with exposure to waters known to be impacted by domestic wastewater.

Table A.2
Recreational Water Quality Criteria for Enterococci Bacteria Concentrations (USEPA 2012)

Pathogen Indicator Bacteria	**Recommendation 1** Estimated Illness Rate 36 per 1,000 recreators		**Recommendation 2** Estimated Illness Rate 32 per 1,000 recreators	
	Geometric Mean (cfu/100mL)	Statistical Threshold Value (cfu/100mL)	Geometric Mean (cfu/100mL)	Statistical Threshold Value (cfu/100mL)
Enterococci	35	130	30	110