



INDICATORS, COLIFORMS AND FECAL STREPTOCOCCI

What is an indicator microorganism?

The number and variety of microbial agents that might be present in domestic wastewater is considerable. The routine monitoring for all the possibilities is either impossible or impractical. The time required to complete most of the analyses precludes their utility as a water quality control feedback tool. The solution to the problem has been the use of indicator bacteria that would be present when potential pathogen containing material (feces) was present. It should be emphasized that the presence of indicator bacteria does not mean the water contains pathogenic microorganisms but rather the potential exists for the presence of pathogens since the indicator bacteria point to the presence of fecal material in the sample. The number of pathogens that might be associated with the concentration of the indicator will be a function of the disease incidence in the community at the time the fecal material was disposed.

The ideal indicator should: 1.) be present only when fecal contamination is present; 2.) exhibit the same or greater survival characteristics as the target pathogen for which it is a surrogate; 3.) not reproduce outside of the host; and, 4.) be readily monitored in a timely manner. At the present time no indicators in common use meet all these criteria.

Bacterial Indicators

The total coliform, fecal coliform and *E. coli* tests have evolved in that order over a period of more than 90 years. The direction of this evolution has been toward a more accurate detection of *E. coli*. This would explain the rather loose accuracy of the total coliform test and the gradual tightening of the measurement as the practice has progressed to the *E. coli* test.

Total Coliform Test

The coliform group includes a number of genera and species of bacteria which have common biochemical and morphological attributes that include gram negative, non-spore forming rods that ferment lactose in 24 to 48 hours at 35C. These attributes are found in *Escherichia coli* which is the coliform of most sanitary significance as it is very common in the feces of warm blooded animals. Historically, the coliform test was developed with the aim to estimate the presence of *E. coli* in water samples by detecting bacteria that had the same cultural attributes. As the test was used it soon became clear that it was not specific for *E. coli*.

and that a variety of bacteria species can be included under the coliform umbrella, many of which are of limited sanitary significance. The total coliform standard is still used in certain jurisdictions (drinking water for example) as it is felt to be a very conservative risk management tool.



Fecal Coliform Test

A subset of the coliform group of bacteria that are able to grow at 44.5C (thermotolerant coliforms). *E. coli* is able to grow at this temperature while most of the coliforms of limited sanitary significance do not. Monitoring methods that employ elevated temperature incubation give a more specific estimate of the presence and number of *E. coli* and thus the presence of fecal contamination. There are some thermotolerant coliform species, such as *Klebsiella*, that are of questionable sanitary significance, thus the test is not an absolute.

E. coli Test

The most recent development in the evolution of the

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coliform test is the ability to specifically identify and quantify *E. coli* in water samples through the use of elevated temperatures and the development of a readily identifiable chemical indicator (MUG) for the presence of an enzyme system specific to *E. coli*. The presence of *E. coli* is a much more certain indication of fecal contamination than is the presence of fecal coliforms. There is a need for an indicator bacterium that could be used to indicate the presence of fecal material as well as indicate the source as human or non-human; unfortunately, *E. coli* does not fill this bill. A great deal of effort continues to be expended to find an indicator that will fill the bill but to date nothing practical has been developed. Differentiation of pathogenic *E. coli* is possible, but not commonly practiced.

Fecal Streptococci Test

The feces of humans and animals contain large numbers of streptococcal bacteria that can be classified as belonging to the fecal streptococci group. There are six species or of streptococci included in this grouping. These streptococci include *S. faecalis*,; *S. faecium*; and, *S. avium* and *S. gallinarum*; *S. bovis*; and, *S. equinus*. In some reports the two subspecies *S. faecalis subsp. liquefaciens*; *S. faecalis subsp. zymogenese* are also described but their validity as subspecies is questionable. Cultural methods analogous to the coliform tests have been developed to determine the presence and concentration of these bacteria in water samples. In the 1950's there was a great deal of interest in these indicator bacteria as they were thought to be only of fecal origin and thus would be more specific than was the total coliform test, which at the time was the only practical test available. This group of bacteria are primarily found only in the feces of warm-blooded animals but it is now understood that some subtypes of this group might be associated with insects. With the advent of the fecal coliform test interest in the use of the fecal streptococci as an important water quality indicator was diminished. The recognition that the fecal streptococci numbers in animal feces was considerably higher than that for fecal coliforms, gave birth to the idea that the

ratio of their numbers relative to each other (FC/FS) in a water sample would be an indication of the source of fecal contamination (human vs animal). It has since been abandoned because of the recognition that the die-off rate of the various species and sub-species of fecal streptococci are quite different, some being even greater than that of the fecal Coliforms.

Enterococci Test

The enterococci are a subset of the fecal streptococci group that include the first four species of fecal streptococci listed above. This group of streptococci are considered to be more specific as indicators of the sanitary quality of recreational waters than the fecal streptococci group as a whole.

Examples of Guidelines and Standards for Indicators in Various Matrices

Matrix	Total Coliform	Fecal Coliform	<i>E. coli</i>	Enterococcus	HPC
Drinking Water	Absent	Absent	Absent		< 500
Bottled Water (Drinking)	Absent	Absent	Absent		<100
Swimming Pool	<1.1				<200
Fresh Bathing	<1000	126		<33	
Ocean Bathing	<240	90% <400; MD <200		35	
Shellfish Growing Waters Approved	< 70 <10%> 230	< 14 <10%> 43			
Restricted	< 700 <10%> 2300	<10%> 60			
Prohibited	> 700				
Shellfish Meat		<230			<500,000
Raw Source Water	20,000 (50-50,000)	2000			
Chlorinated Sewage Effluents	2.2-230		<126	<33	

The counts in all water samples are per 100 mL; Shellfish meat counts are per 100 grams.