

APPENDIX J. COLIFORM CHARACTERISTICS AND ANALYSIS METHODS

Coliform bacteria are commonly divided into two categories based on controlled growth in the laboratory. Total Coliform is a large group of bacteria that belong to the family Enterobacteriaceae and includes all aerobic and facultative anaerobic, gram-negative, nonspore-forming, rod shaped bacteria that ferment lactose with gas formation within 48 hours at 35°C (APHA, 1998).

The two standard methods for coliform analyses are , multiple tube fermentation, where replicate tubes and dilutions are reported in terms of the most probable number (MPN) and the direct plating technique of membrane filtration (MF) (APHA, 1998). In either case, results are reported relative to a 100-mL sample. Results of multiple tube fermentation tests are reported in terms of the most probable number (MPN) which is determined from probability formulas that estimate the average density of coliform bacteria in a sample based on the results of a number of dilutions from a single sample (APHA, 1998). MF results are based on colony counts and are reported relative to the 100-mL sample.

Results from MPN analyses may differ considerably from results derived from MF methodologies for a number of reasons. The two most commonly reported reasons being, (1) the multiple tube fermentation media and incubation temperature allows non-target bacteria from other groups to grow and (2) the media used in multiple tube fermentation analyses revives stressed or injured organisms that would not be enumerated by MF techniques (Griffin, 2004). Although MF methodologies may result in a lower number of bacteria reported, the results are highly reproducible. Constraints with MF techniques include poor results with turbid water samples and the growth of non-target bacteria on the growing medium, which results in samples where the number of colonies grown cannot accurately be determined. The presence of large quantities of algal cells can interfere with MF analyses, making MPN methodologies more representative for some natural waters (APHA, 1998).

The Class III Fresh Water State Standards for coliforms reported in coliform forming units (cfu) that applies to Gainesville's urban creeks are as follows:

Fecal counts shall not exceed a monthly average of 200 cfu / 100 mL, nor exceed 400 cfu / 100 mL in 10% of the samples, nor exceed 800 cfu / 100 mL on any one day. Monthly averages shall be expressed as geometric means based on a minimum of 10 samples taken over a 30 day period. (Chapter 62-302, F.A.C., FDEP 2002)

Total coliform counts shall be $\leq 1,000$ cfu / 100 mL as a monthly average; $\leq 1,000$ cfu / 100 mL in 20% of the samples examined during any month; $\leq 2,400$ at anytime. Monthly averages shall be expressed as geometric means based on a minimum of 10 samples taken over a 30 day period, using either MPN or MF counts (Chapter 62-302, F.A.C., FDEP 2002).