



## Certification Requirements for the Total Coliform/*E.coli* Enzyme Substrate Test Procedures

The following documentation is required to become certified to perform one of the Enzyme Substrate test procedures:

- 1) A completed application form with the application fee.
- 2) A Standard Operating Procedure (SOP) for performing the analysis. Refer to the EPA's Manual for the Certification of Laboratories Analyzing Drinking Water, Fifth Edition EPA 815-R-05-004 for Criteria and Procedures, Quality Assurance. The requirements in this manual must be included in the SOP in addition to the quality control discussed at the bottom of the page. Ensure that the items below are also addressed in the SOP.
  - a) The wattage of the long wave fluorescent ultraviolet lamp (must be at least six watts).
  - b) The water supply used must be sterilized deionized or distilled (not buffered dilution water) produced in-house or purchased from a laboratory supply vendor. Reagent water quality control checks listed in the EPA's Drinking Water Manual do not have to be performed if the water used is of the quality mentioned above.
  - c) The collection/culture vessel (must be borosilicate glass or equivalent and cannot autofluoresce).
  - d) The use and active life of the color/fluorescence comparator.
- 3) Acceptable proficiency testing (PT) sample results from an A2LA approved PT provider.
- 4) Complete quality control and analysis records for the PT sample.

Any laboratory performing the test procedure must check each lot of medium received into the laboratory by using known cultures of microorganisms before that lot is used for compliance monitoring purposes. Laboratories should use the culture organisms recommended by the medium manufacturer. Typical organisms include: *Escherichia coli* as the positive control, *Pseudomonas aeruginosa* as the negative control, and *Klebsiella pneumoniae* for the total coliform positive/*E.coli* negative control (positive color produced but without fluorescence). Sterile deionized/distilled water must be pipetted into a single vessel of medium and incubated as a sterility control.

Laboratories must follow the manufacturer's storage requirements, including expiration dates, for each lot of medium and comparator received into the laboratory.

On a weekly basis, or each time that the medium is used if used less than once each week, sterile deionized/distilled water must be pipetted into one vessel of the medium, inoculated with growth from a living agar slant of *Escherichia coli* or a known positive sample may be incubated (stream, creek, wastewater) and incubated as a known positive control. On a weekly basis or each time that the medium is used if used less than once each week, sterile deionized or distilled water must be pipetted into a single vessel of the medium and incubated as a sterility control.

If you have questions about the certification requirements for Total Coliform/*E.coli* Enzyme Substrate test procedures, please contact the Office of Environmental Laboratory Certification at (803) 896-0970.