



Certification Requirements for Enzyme Substrate Test Procedures for Total Coliform/*E.coli*, *E. coli*, and Fecal Coliforms

The following documents certification requirements to perform one of the Enzyme Substrate test procedures using media such as Colilert, Colilert-18, Colitag, Colisure, etc. An application along with the required documentation must be submitted.

The application and application checklist are located:

<http://www.scdhec.gov/environment/EnvironmentalLabCertification/ELCHowToApply/>

A list of required documentation is located:

<http://www.scdhec.gov/Environment/EnvironmentalLabCertification/RequiredDocumentation/>.

- The wattage of the long wave fluorescent ultraviolet lamp must be at least six watts. Upon purchase the output of the UV lamp must be determined with an UV light meter, or, alternatively, a new UV bulb must be put into service every year. (Not required for fecal coliform only testing)
- The reagent water used must be sterilized non-buffered deionized or distilled water produced in-house or purchased from a commercial vendor.
- The culture vessel (must be borosilicate glass or equivalent and cannot auto fluoresce). Typically, plastic bottles are used instead of glass containers. (Autofluorescence check not required for fecal coliform only testing)
- A color/fluorescence comparator is required for Presence-Absence methods and enumeration methods when offered by the manufacturer.
- Complete quality control and analysis records for the PT sample.
- Laboratories must follow the manufacturer's storage requirements, including expiration dates, for each lot of medium and comparator received into the laboratory.
- Any laboratory performing the test procedure must check the quality control (QC) of each lot of medium received into the laboratory by using known purchased control cultures of microorganisms and a sterility control before that lot can be used for compliance monitoring purposes and every 90 days thereafter. Laboratories should use the control culture organisms recommended by the medium manufacturer. These control cultures must be commercially purchased and the Certificates of Analysis must be maintained. Typical organisms include: *Escherichia coli* (*E. coli*) as the positive control, *Pseudomonas aeruginosa* as the negative control, and *Klebsiella pneumoniae* or *K. oxytoca* for the total coliform positive/*E.coli* negative control (positive color produced but without fluorescence). An *E. coli* positive control and a negative fecal coliform control such as *P. aeruginosa* are required for fecal coliforms. The sterility control sample is one packet of the medium added to 100 mL of sterile deionized or distilled water.
- Each week that samples are analyzed, an *E. coli* positive control sample (or fecal coliform positive for fecal coliform only testing) and a sterility control sample must be analyzed. The weekly positive control sample consists of one packet of the medium added to 100 mL of sterile deionized or distilled water and then inoculated with an *E. coli* containing QC sample or stock (fecal coliform positive is required for fecal coliform only testing). Alternatively, a stream sample, influent sample, effluent sample, etc., that is known to be *E. coli* positive (or other fecal coliform positive for fecal coliform only testing) can be analyzed as the weekly known positive sample.

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