

General Information

Contents: 20 test (Vial format)

One vial is sufficient for a 100 ml water sample.

Application

Presence/Absence test for the simultaneous detection of total coliforms and E.coli, both of which have to be monitored under USEPA Total Coliforms Rule (Sec. 141.21)

Principle

The high nutritional quality of the peptones and the incorporated phosphate buffer support rapid growth of coliforms. Laurylsulphate largely inhibits the accompanying flora, especially gram-positive organism. The simultaneous detection of coliforms and E.coli is made possible by adding chromogenic substrate X-GAL and the fluorogenic substrate MUG. A color change to blue-green in the broth indicates the presence of total coliforms. E.coli additionally creates a blue fluorescence under UV-light (365 nm).

Composition in g / tube pack

Tryptose 0.5; sodium chloride; sorbitol 0.1; tryptophan 0.1; di-potassium hydrogen phosphate 0.27; potassium dihydrogen phosphate 0.2; Laurylsulphate sodium salt 0.01; X-GAL 0.008; MUG 0.005; IPTG 0.01

Procedure

Note: In case you cannot start testing the water sample right away. Test has to be done

- Within 6 hours after receiving the water sample if stored at room temperature
- Within 24 hours if stored in the refrigerator.

1. Take one tube; lightly tap to ensure the granules are at the bottom. Screw the cap until it opens.

DO NOT TOUCH THE OPENING TO AVOID CONTAMINATION!

2. Add the content to a 100 ml water sample in a sterile, transparent non-flourescing vessel with a minimum capacity of 120 ml.
3. Shake the capped vessel to dissolve granules completely.
4. Incubate the vessel at 35-37 °C for up to 24 hours at 20-25 °C for up to 48 hours.

Interpretation of Results

No color change (the broth remains yellowish in color): absence of coliforms and E.coli.

Color change to blue-green

Total coliforms:

Any color change of the broth to blue-green, even if only in the upper section, confirms the presence of total coliforms (X-GAL reaction).

E.coli: Look for fluorescence in blue-green colored vessel using UV light (365nm) in front of the vessel (toward the the sample from your eyes). /a bluish fluorescence indicates presence of E.coli (MUG reaction).

Attention: Protect eyes from UV light

Confirmation of E.coli takes 30 seconds to complete:

There are other species of bacteria able to create a positive fluorescence with MUG. The simple indole reaction distinguishes E.coli from other microbes! To conform E.coli in the vessel with positive fluorescence overlay the broth with KOVAC's reagent.

A red ring confirms the presence of E.coli.

	Color change to blue green	Fluorescence	Confirmation: Indole reaction
Total coliforms	+	-	-
E.coli	+	+	+
Negative	No color change		

Quality Control

Test strain	Growth	Color change	Fluorescence	Indole Reaction
E.coli ATCC 11775	+	+	+	+
Citrobacter ATCC 8090	+	+	-	-
Salmonella typhimurium ATCC 14028	+	-	-	-

Disposal

Autoclave the broth (15 min/121 °C).

Alternatively heat the broth for 30 min. in boiling water or use a proper disinfectant.

Storage

Store dry at 15 °C to 25 °C.

Shelf-life

If stored under recommended conditions the unopened snap pack has a shelf-life of 3 years after day of production (see expiry date on the label).