

The Rapid Hicoliform™ Test Kit is used for detection and confirmation of *Escherichia coli* and total coliforms on the basis of enzyme substrate reaction from water samples, using a combination of chromogenic and fluorogenic substrate.

Composition :

Ingredients	Gms/pack
Peptone, special	0.50
Sodium chloride	0.50
Sorbitol	0.1
Dipotassium hydrogen phosphate	0.27
Potassium dihydrogen phosphate	0.2
Sodium lauryl sulphate	0.01
Chromogenic substrate	0.008
Fluorogenic substrate	0.005
Isopropyl- β -D-thiogalactopyranoside (IPTG)	0.01

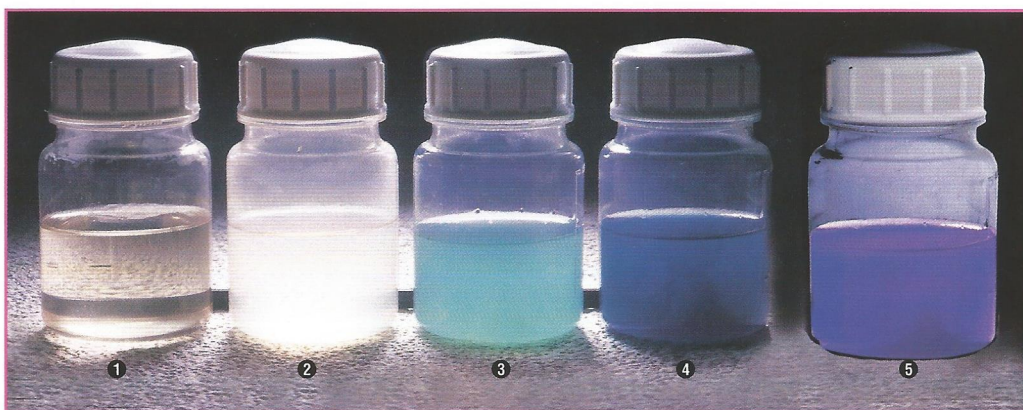
Direction :

Collect 100 ml water to be tested in sterile disposable bottle. Add entire quantity of medium by swirling to dissolve the powder completely. After dissolution, incubate the bottle for 24-48 hours at 35-37°C. Observe the colour change of the

medium from light yellow to blue green indicating the presence of coliforms and light yellow to fluorescent blue green (under uv) indicating presence of *Escherichia coli*.

Principle and Interpretation :

The rapid Hicoliform™ Test Kit is used for the simultaneous detection of total coliforms and *E. coli*. Peptone, special which is rich in tryptophan content, provides essential growth nutrients and is useful for the simultaneous detection of indole production. The presence of indole can be detected by addition of p-dimethylaminobenzaldehyde indicated by formation of red coloured ring. Sorbitol provides the carbon source. The phosphate salts provide buffering action for rapid growth of coliforms. Sodium lauryl sulphate makes the medium selective by inhibiting accompanying microflora, especially the gram-positive organisms. The fluorogenic substrate, is split by enzyme β -D-glucuronidase, which is specifically found in *E. coli*. The reaction is indicated by a blue fluorescence under UV light. The presence of total coliforms is indicated by a blue-green colour of the broth due to cleavage of chromogenic substrate. IPTG amplifies enzyme synthesis and increases the activity of β -D-galactosidase.



1. Control 2. *S. Typhimurium* (Negative reaction) 3. Total coliforms (Positive reaction) 4. *E. coli* (Positive reaction) 5. *E. coli* Fluorescence under UV (Positive reaction with fluorescence)

Quality Control :

Appearance:

Light yellow to yellowish brown coloured, homogeneous, free flowing powder.

Colour and Clarity :

Light yellow coloured, clear solution.

Cultural Response :

Cultural characteristics observed after an incubation of 24–48 hours at 35-37°C.

Organism	Colour change in medium	Fluorescence	Indole reaction
Total coliforms	blue-green*	—	—
<i>E. coli</i> (25922)	blue-green*	+	+
<i>S. Typhimurium</i> (23564)	yellow**	—	—
<i>S. Typhimurium</i> (14028)	yellow**	—	—

Key: * = Positive reaction – colour change to blue green

** = Negative reaction – no colour change (pale yellow)

References :

- Hahn, G., and Wittrock E. 1991, *Acta Microbiologica Hungarica* 38(3-4):265-271.
- Manafi. M., and Kneifel, W. 1989. *Zbl. Hygiene and Umweltmedizin* 189:225-234.
- Manafi, M. 1990. *Forum Stadte-Hygiene* 41:181-184.
- Manafi, M. 1991. *Ernahrung / Nutrition*, 15, Nr. 10.
- Manafi, M., and Kneifel, W. 1991, *Acta Microbiologica Hungarica* 38(3-4):293-304.
- Manafi, M., Kneifel B., and Bascon, S. 1991. *Microbiol. Rev.* 55:335-348.

Storage and Shelf-life :

On receipt store between 2 - 8°C. It has shelf-life of 3 years.