

### **Tryptone Water (i23185)**

Tryptone Water is used for cultivating non-fastidious organisms, for studying carbohydrate fermentation patterns and for performing the indole test.

#### **Principles & Uses**

Tryptone Water, a recommended medium for the detection of *Enterobacteriaceae*, particularly *E. coli*, in water and food samples based on indole production, plays a pivotal role in bacterial classification and identification. It's endorsed by various institutions like APHA and ISO committees for assessing indole production, a key feature used in differentiation. Peptone, rich in nitrogen, vitamins, minerals, and essential amino acids, serves as an ideal substrate for indole production due to its high tryptophan content. Certain bacteria employ hydrolytic enzymes to break down tryptophan, resulting in indole formation, a process detectable with Kovacs Indole or Ehrlich's Reagent.

The red coloration that forms when indole combines with the reagent's aldehyde is a visual indicator. Tryptone Water, in conjunction with Brilliant Green Bile Broth 2%, allows the determination of the most probable number (MPN) of *E. coli* in food samples, based on growth, gas production in M121, and indole production in Tryptone Water. This aids in conducting the presumptive *E. coli* test. Although indole testing aids in microbial differentiation, complete organism identification necessitates further biochemical confirmation.

### Composition (gr/L)

Peptone 10, Sodium chloride 5. Final pH at 25°C 7.3 ± 0.2

### **Preparation from dehydrated Powder**

Dissolve 15 g of the powder in 1 Liter of purified water. Autoclave at 121°C for 15 minutes.

# For Determining Carbohydrate Fermentation Patterns:

Add 1.8 mL 1% phenol red solution to 1 Liter rehydrated Peptone Water. Mix thoroughly. Dispense into test tubes containing inverted Durham vials. Autoclave at 121°C for 15 minutes. Aseptically add sufficient sterile carbohydrate solution to yield a 1% final concentration. Rotate each tube to thoroughly distribute the carbohydrate.

### **Quality Control**

Dehydrated Appearance: White to light beige, free flowing, homogeneous.

Prepared Appearance: Light to medium amber, clear to slightly opalescent.

Reaction of 1.5% Solution at 25°C: pH 7.3 ± 0.2

### **Cultural Response**

Inoculate and incubate at  $35 \pm 2^{\circ}$ C for 18-24 hours. Add 0.5 mL Indole Reagent (Kovacs) to the tubes to test for indole production. Formation of a red color denotes a positive indole test.

Organism (ATCC*)	Recovery	Indole Production
Escherichia coli (25922)	Good	+
Enterobacter cloacae (13047)	Good	-

\*ATCC is a registered trade mark of the American Type Culture Collection.



E. coli with a Durham tube filled with gas (left). E. coli with a red ring indicates a positive indole test (right).



## Storage

Keep the container at 15-30 °C. Store prepared medium at 2-8 °C.