

Technical Data

Tryptone Phosphate Broth

Tryptone Phosphate Broth is recommended for enrichment and cultivation of enteropathogenic *Escherichia coli* from suspected food samples.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	20.000
Dipotassium phosphate	2.000
Monopotassium phosphate	2.000
Sodium chloride	5.000
Polysorbate 80	1.500
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 30.5 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Dispense in 100 ml aliquotes and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Certain biotypes of *Escherichia coli* are etiological agents of gastrointestinal illness in humans and several mammals (1). These enteric diseases are characterized by fever, vomitting and prominent and watery diarrhea, usually with mucus but not blood. Enteropathogenic *E. coli* serotypes have been shown to be important causes of infantile diarrhoea (2). Tryptone Phosphate Broth is formulated as recommended by APHA (3) for the enrichment of Enteropathogenic *E. coli*.

Casein enzymic hydrolysate serves as a good source of nitrogen. Polysorbate 80 is the fatty acid source required for bacterial metabolism. The inorganic phosphates serve as the buffer while sodium chloride maintains the osmotic balance.

Examine test samples as promptly as possible after receipt. Refrigerate perishable material less than 1 day to avoid damages to the bacteria. Aseptically weigh 25 grams test portion into 225 ml Brain Heart Infusion Broth (M210). Agitate gently, and incubate for 2 hours at 35°C. After incubation, streak loopful on MacConkey Agar (M081) and on EMB Agar (M022). Incubate at 35°C for 2 hours. Pour the supernatant into 250 ml double strength Tryptone Phosphate Broth. Incubate at 44°C for 20 ± 2 hours. Subsequently streak on EMB Agar (M022) and MacConkey Agar (M081). Tryptone Phosphate Broth helps to enrich the stressed bacteria, if present.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light amber coloured clear solution without any precipitate.

Reaction

Reaction of 3.05% w/v aqueous solution at 25°C. pH : 7.0 \pm 0.2

pН

6.80-7.20

Cultural Response

M953: Cultural characteristics observed after an incubation at 44°C for 18-24 hours.

Organism	Inoculum	Growth
	(CFU)	
Escherichia coli ATCC	50-100	good-luxuriant
25922		

M953

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1. Levin M. M., 1987, J. Infect. Dis. 155: 377

2. Murray P. R., Baron J. H., Pfaller M. A., Jorgensen J. H. and Yolken R. H., (Ed.), 2003, Manual of Clinical Microbiology, 8th Ed., American Society for Microbiology, Washington, D.C.

3. Downes F. P. and Ito K., (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C.

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