



Buffered Tryptone Glucose Yeast Extract Broth

M951

Intended Use:

Recommended for cultivation and characterization of Clostridia isolated from food specimens.

Composition**

Ingredients	Gms / Litre
Tryptone	50.000
Peptone	5.000
Yeast extract	20.000
Dextrose (Glucose)	4.000
Disodium hydrogen phosphate	5.000
Sodium thioglycollate	1.000
Final pH (at 25°C)	7.3±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 8.5 grams in 100 ml purified / distilled water. Heat, if necessary to dissolve the medium completely and dispense 15 ml into 20x150 mm test tubes or 100 ml in 170 ml bottles. Sterilize by autoclaving at 15 lbs pressure (121°C) for 8 minutes (tubes) or 15 minutes (bottles).

Principle And Interpretation

Clostridium perfringens food poisoning is one of the most common types of foodborne illness (3). A heat labile enterotoxin produced only by sporulating cells (4) induces the major symptoms of diarrhoea in perfringens poisoning (1). Although the enterotoxin is not preformed in the food, the foods in which conditions are favorable for sporulation may contain enterotoxin (2,7). Buffered Tryptone Glucose Yeast Extract Broth is prepared as recommended by APHA (8) for enrichment as well as for cultivation of Clostridia from food samples. Buffered Tryptone Glucose Yeast Extract Broth is also used to obtain pure cultures of *Clostridia* before proceeding for confirmation. Endospores are not usually produced in this medium(8).

The medium contains tryptone, peptone and yeast extract, which provides carbon and nitrogen, vitamins and other essential nutrients. Dextrose is the fermentable sugar. Disodium phosphate buffers the medium well. Sodium thioglycollate present in the medium acts as a reducing agent and maintains a low oxygen tension in the medium.

Type of specimen

Food samples

Specimen Collection and Handling

For enrichment 2 grams of food sample is inoculated in 15-20 ml of sterile Buffered Tryptone Glucose Yeast Extract Broth (M951). After incubation of 20-24 hours at 35-37°C the culture from the tubes shows turbidity. After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

- 1. Further recovery on selective media is necessary to obtain pure colonies of culture.
- 2. Biochemical and serological testing of pure colony is required for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

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 8.5% w/v aqueous solution at 25°C. pH : 7.3±0.2

pН

7.10-7.50

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth
<i>Clostridium botulinum</i> ATCC 25763	50-100	good-luxuriant
<i>Clostridium perfringens</i> ATCC 12924	50-100	good-luxuriant
<i>Clostridium sporogenes</i> ATCC 11437	50-100	good-luxuriant

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

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User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (5,6).

Reference

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- 4. Duncan C. L., 1973, J. Bacteriol., 113:932.
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6. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

7. Naik M. S., and Duncan C. L., 1977, J. Food Safety, 1:7.

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