Tryptone Bile Agar

Intended Use
Tryptone Bile Agar is used for rapid detection and enumeration of *Escherichia coli* in food using a modified direct plating method.

Summary
Tryptone Bile Agar was formulated by Anderson and Baird-Parker. The International Commission on the Microbiological Specifications for Foods (CMSF) compared the Most Probable Number (MPN) and the Anderson-Baird-Parker Direct Plating Method (DPM) and observed that DPM was superior to MPN for enumeration of *Escherichia coli* from raw meats. Superiority of DPM method was noticed by the organization on the basis of less variability, better recovery from frozen samples, greater rapidity and the smaller quantity of medium required. The DPM enumerates both anaerogenic and late lactose fermenting strains of *E. coli* which could be missed by the MPN method (about 10%). This formulation is recommended by ISO committee for the enumeration of *E. coli*. Holbrook et al modified the DPM for detection and enumeration of sublethally damaged cells of *E. coli* in frozen, dried, heat processed or acid foods and found that resuscitation step reduces the high concentration of sugar present in the inoculum to a level which does not interfere with the production of indole as the synthesis of tryptophanase is inhibited by high sugar concentrations.

Principle
Certain organisms breakdown the amino acid tryptophan with the help of enzymes that mediate the production of indole by hydrolytic activity. The indole produced can be detected by either Kovac's or Ehrlich's reagent. Indole combines with the aldehyde present in the above reagent to give red colour in the alcohol layer. The alcohol layer extracts and concentrates the red colour complex. The indole positive organisms other than *E. coli* are inhibited by bile salts and elevated incubation temperature.

Formula*

Ingredients	**g/L**
Casein enzymic hydrolysate	20.0
Bile salt mixture	1.5
Agar	15.0
Final pH (at 25°C)	7.2 ± 0.2

*Adjusted to suit performance parameters

Storage and Stability
Store dehydrated medium below 30°C in tightly closed container and the prepared medium at 2°C-8°C. Avoid freezing and overheating. Use before expiry date on the label. Once opened keep powdered medium closed to avoid hydration.

Type of Specimen
Food sample

Specimen Collection and Handling
Ensure that all samples are properly labelled.
Follow appropriate techniques for handling samples as per established guidelines.
Some samples may require special handling, such as immediate refrigeration or protection from light, follow the standard procedure.
The samples must be stored and tested within the permissible time duration.
After use, contaminated materials must be sterilized by autoclaving before discarding.

Directions
1. Suspend 36.50 g of the powder in 1000 mL purified / distilled water.
2. Heat to boiling to dissolve the powder completely.
3. Sterilize by autoclaving at 121°C (15 psi) for 15 minutes as per validated cycle.
4. Mix well and dispense as desired.

Quality Control

Dehydrated Appearance: Cream to yellow coloured , homogenous, free flowing powder
Prepared Appearance: Yellow coloured, clear to slightly opalescent gel forms in petridishes.
Cultural Response: Cultural characteristics observed after an incubation of 24 hours at 44°C.

Organism (ATCC)	Growth
Escherichia coli (25922)	Good
Klebsiella aerogenes (13048)	Partial Inhibition

Performance and Evaluation

Performance of the product is dependent on following parameters as per product label claim:

1. Directions
2. Storage
3. Expiry

Warranty

This product is designed to perform as described on the label and package insert. The manufacturer disclaims any implied warranty of use and sale for any other purpose.

References

1. Anderson J. M. and Baird-Parker A. C., 1975, J. Appl. Bacteriol., 39:111.
2. International Commission on Microbiological Specifications for Food, 1979, Can. J. Microbiol., 25:1321.
3. Ewing W. H., 1972, US Dept. of Health, Education and Welfare, CRC, Atlanta.
4. International Organization for Standardization (ISO), 1988, Draft ISO/DIS 6391.
5. Holbrook R., Anderson J. M. and Baird - Parker A.C., 1980, Food Technol. in Aust., 32:78.
6. Clarke P. H. and Cowen S. T., 1952, J. Gen. Microbiol., 6:187.
7. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
8. Finegold S. M., Baron E. J., 1986, Bailey and Scotts Diagnostic Microbiology, 7th Ed., The C.V. Mosby Co., St. Louis.
9. Data on file: Microxpress®, A Division of Tulip Diagnostics (P) Ltd.

Product Presentation:

Cat No.	Product description	Pack Size
201200240100	Dehydrated culture Media	100 g
201200240500	Dehydrated Culture Media	500 g