

08069 Modified Tryptone Soya Broth (Modified Soyabean Bile Broth Base; Tryptic Soy Broth, modified; mTSB)

Modified tryptone soy broth is recommended as an enrichment medium for the detection of *Escherichia coli* O157:H7 from food and animal feeding stuffs.

Composition:

Ingredients	Grams/Litre
Casein enzymic hydrolysate	17.0
Papaic digest of soyabean meal	3.0
D-Glucose	2.5
Bile salts mixture	1.5
Dipotassium hydrogen phosphate	4.0
Sodium chloride	5.0
Final pH (at 25°C) 7.3 ± 0.2	

Store prepared media below 8°C, protected from direct light. Store dehydrated powder, in a dry place, in tightly-sealed containers at 2-25°C.

Appearance: Faintly beige coloured, homogeneous, free flowing powder.

Color and Clarity: Light yellow coloured, clear solution.

Directions:

Suspend 33 grams of Modified Tryptone Soy Broth in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121 °C) for 15 minutes. Cool to 45-50 °C and aseptically add rehydrated contents of 1 vial of VCC Selective Supplement (Cat. No. 80704). Mix well and dispense as desired.

Principle and Interpretation:

Escherichia coli is commonly found in the gut of humans and warm-blooded animals. In humans, it is found in several intestinal and extra-intestinal infections. Based on the virulence properties, different strains are divided into Enterotoxigenic (ETEC), Enteropathogenic (EPEC), Enteroinvasive (EIEC), Verotoxigenic (VTEC) and Enterohaemorrhagic (EHEC). *Escherichia coli* O157:H7 is an Enterohaemorrhagic strain implicated in foodborne illness.

Due to severe nature of the clinical symptoms and the high contagiousness of the pathogens, the detection of EHEC is

constantly gaining more and more clinical relevance (1, 2).

Modified Soyabean Bile Broth is formulated as recommended by FDA (3) for the enrichment and isolation of *E. coli* O157:H7. Modified Soyabean Bile Broth was subsequently accepted by ISO Committee as an enrichment medium for the detection and isolation of *E. coli* O157:H7 under the specification ISO/DIS 16654: 1999 (4).

Casein enzymic hydrolysate and papaic digest of soyabean meal provide carbonaceous, nitrogenous compounds and other essential growth nutrients. D-Glucose is the fermentable carbohydrate and energy source. Bile salts mixture inhibits gram-positive bacteria. Sodium chloride maintains osmotic equilibrium while phosphate buffers the medium well. Novobiocin renders the medium selectivity. Whenever low levels of *E. coli* O157:H7 are suspected, the food is enriched in Modified Soyabean Bile Broth and further plated on selective medium as Sorbitol MacConkey Agar (Cat. No. 88902) for isolation and identification.

Blend 25 grams food sample to be tested in 224 ml Modified Soyabean Bile Broth and incubate with shaking (about 100 rpm) at 37°C for 18-24 hours. Prepare dilution of the enrichment culture with phosphate buffer and spread 0.1 ml of each dilution on HC Agar plates and incubate at 43°C for 24 hours.



Cultural characteristics after 18-24 hours at 35-37°C.

Organisms (ATCC)	Inoculum [cfu]	Growth on Sorbitol MacConkey Agar	Recovery
<i>Escherichia coli</i> O157:H7	50-100	+++	≥50%
<i>Escherichia coli</i> (25922)	50-100	++	≥40%
<i>Enterococcus faecalis</i> (29212)	≥10 ³	-	0%
<i>Staphylococcus aureus</i> (25923)	≥10 ³	-	0%

References:

1. R.A. Szabo, E.C. Todd, A. Ean, Method to isolate *E. coli* O157:H7 from food, J. Food Prot., 10, 768-772 (1986)
2. M.A. Karmali, M. Petric, C. Lim et al, J. Infect. Dis., 151, 775 (1985)
3. FDA Bacteriological Analytical Manual, 18th Ed., AOAC, Washington, D.C. (2005)
4. International Organization for Standardization (ISO), Draft ISO/DIS 16654 (1999)

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

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