



## Soyabean Bile Broth w/ Novobiocin

M1654

Soyabean Bile Broth w/ Novobiocin is an enrichment Medium for the detection of *Escherichia coli O157:H7* from food. It is recommended by ISO Committee under the specification ISO/DIS 16654:1999.

### Composition\*\*

Ingredients	Gms / Litre
Casein enzymic hydrolysate	17.000
Papaic digest of soyabean meal	3.000
D-Glucose	2.500
Sodium chloride	5.000
Dipotassium hydrogen phosphate	4.000
Bile salts mixture	1.500
Novobiocin	0.020
Final pH ( at 25°C)	7.3±0.2

\*\*Formula adjusted, standardized to suit performance parameters

### Directions

Suspend 33.02 grams 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.

### Principle And Interpretation

Soyabean Bile Broth is formulated as recommended in FDA (a), for the enrichment and isolation of *Escherichia coli O15:H7* and subsequently accepted by ISO Committee as an enrichment medium for the detection and isolation of *Escherichia coli O15:H7* under the specification ISO/DIS 16654 :1999 (2).

Casein enzymic hydrolysate, papaic digest of soyabean meal provide carbonaceous, nitrogenous compounds and other essential growth nutrients. Dextrose 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 *Escherichia coli O157:H7* are suspected, the food is enriched in Soyabean Bile Broth w/Novobiocin and further plated on selective medium as Sorbitol MacConkey Agar (M298I) or Hemorrhagic coli (HC) Agar (M1158) for isolation and identification.

Blend 25 grams food sample to be tested in 225 ml Soyabean Bile Broth w/Novobiocin and incubate with shaking (about 100 rpm) for 18-24 hours at 41.5°C ± 1. 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.

### Quality Control

#### Appearance

Cream to yellow coloured homogeneous free flowing powder

#### Colour and Clarity of prepared medium

Light yellow coloured clear solution

#### Reaction

Reaction of 1.65% w/v aqueous solution at 25°C. pH : 7.3±0.2

#### pH

7.10-7.50

#### Cultural Response

M1654: Cultural characteristics observed after an incubation at 35-37°C or 41.5±1°C for 18-24 hours.

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Organism	Inoculum (CFU)	Growth
<b>Cultural Response</b>		
<i>Escherichia coli</i> O157 : H7	50-100	luxuriant
<i>Escherichia coli</i> ATCC 25922	50-100	luxuriant
<i>Enterococcus faecalis</i> ATCC 29212	$\geq 10^3$	inhibited
<i>Staphylococcus aureus</i> ATCC 25923	$\geq 10^3$	inhibited

### Storage and Shelf Life

Store dehydrated powder and prepared medium at 2-8°C. Use before expiry period on the label.

### Reference

1. Bacteriological Analytical Manual, 1995, 8th ed., Food and Drug Administration, AOAC International, Gaithersburg, USA.
2. International Organization for Standardization (ISO), 1999, Draft ISO/DIS 16654.

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