

Modified Tryptone-Soy Broth (NCM0196)

Intended Use

Modified Tryptone-Soy Broth has emerged as the medium of choice for the enrichment of *E. coli* O157:H7 in red meats and is incorporated into ISO/TS 13136:2012. This medium is not intended for use in the diagnosis of disease or other conditions in humans.

Description

Concern regarding *E. coli* O157:H7 has grown due to the severity of the disease syndromes caused, and the increase in foodborne infection, so too has the need to optimize methods for its efficient isolation. Symptoms start with severe stomach cramps and watery, bloody diarrhea, and a percentage of individuals infected will develop Haemolytic Uraemic Syndrome (HUS) leading to acute renal failure. In a comparison of four different selective broth media, MTSB was the most productive and selective for the isolation of *E. coli* O157:H7. mTSB is made selective for O157:H7 by including bile salts in the dehydrated medium, and the addition of novobiocin supplement (X150).

Typical Formulation

Enzymatic Digest of Casein	17.0 g/L
Sodium Chloride	5.0 g/L
Dipotassium Hydrogenphosphate	4.0 g/L
D(+)-Glucose	2.5 g/L
Enzymatic Digest of Soy	3.0 g/L
Bile Salts No.3	1.5 g/L

Final pH: 7.2 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Supplements

X150 Novobiocin – 1 vial per 500mL

Precaution

Refer to SDS

Preparation

1. Dissolve 33 grams of the powder in one liter of purified water.
2. Heat with frequent agitation to completely dissolve the medium if necessary.
3. Autoclave at 121°C for 15 minutes.
4. Cool to 45-50°C and add 2 vials of Novobiocin supplement X150.

Test Procedure

Add 25g sample to 225mL of supplemented MTSB and homogenize for 2 minutes. 42°C aerobically for 24hrs. Subculture onto CT-SMAC (NCM0167) or SMAC-BCIG (NCM1007) and examine for non-sorbitol fermenting colonies and/or glucuronidase negative organisms. Some microbiologists recommend the use of an immunomagnetic separation step after 6hrs incubation. Please see our Captivate™ range for further details.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing and beige.

Prepared Appearance: Prepared medium is a clear, pale yellow liquid.



Technical Specification Sheet



Minimum QC:

Escherichia coli O157:H7 (non-toxigenic) WDCM 00014
Escherichia coli WDCM 00013

Results

Turbidity in the broth indicates growth. All broths should be sub-cultured to selective media whether turbid or not.

Expiration

Refer to expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing or appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedures

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Storage

Store dehydrated culture media at 2-30°C away from direct sunlight. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light by keeping container tightly closed.

References

1. Microbiology of food and animal feed - Real-time polymerase chain reaction (PCR)-based method for the detection of foodborne pathogens - Horizontal method for the detection of Shiga toxin-producing *Escherichia coli* (STEC) and the determination of O157, O111, O26, O103 and O145 serogroups (ISO/TS13136:2012).
2. Bolton, E.J., Crozier, L., Williamson, J.K. (1995) Optimisation of methods for the isolation of *Escherichia coli* O157 from beef burgers. PHLS Microbiology Digest 12 (2) 67-70.
3. Willshaw, G.A., Smith, H.R., Roberts, D., Thirlwell, J., Cheasty, T., Rowe, B. (1993) Examination of raw beef products for the presence of verocytotoxin producing *Escherichia coli*, particularly those of serogroup O157. J.Appl.Bacteriol. 75 420-426.
4. Sharp, J.C.M., Coia, J.E., Curnow, J., Reilly, W.J. (1994) *Escherichia coli* O157 infections in Scotland. J.Med.Microbiol 40 3-9.
5. Doyle, M.P. (1991) *Escherichia coli* O157:H7 and its significance in foods. Int.J.Food Microbiol. 12 289-302.



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