Tryptic Soy Broth, Modified with Acid Digest of Casein (NCM0026)

Revision: 1 Effective Date: 2/5/2021

Intended Use

Tryptic Soy Broth, Modified with Acid Digest of Casein is used for the selective enrichment of enterohemorrhagic *E. coli* in foods in a laboratory setting. Tryptic Soy Broth, Modified with Acid Digest of Casein is not intended for use in the diagnosis of disease or other conditions in humans. This medium conforms to USDA Formulation with Acid Digest of Casein.

Description

The first major outbreak of *E. coli* O157:H7 was in 1982 and traced to contaminated hamburgers. Other known sources of infection include sprouts, lettuce, salami, unpasteurized milk, juice and/or swimming in or drinking contaminated water. Tryptic Soy Broth, Modified with Acid Digest of Casein is used to enrich food samples suspected of having low levels of EHEC during pathogen testing.

Typical Formulation

Enzymatic Digest of Casein	17.0 g/L
Acid Digest of Casein	10.0 g/L
Sodium Chloride	5.0 g/L
Dipotassium Phosphate	4.0 g/L
Enzymatic Digest of Soybean Meal	3.0 g/L
Dextrose	2.5 g/L
Bile Salts No. 3	1.5 g/L

Final pH: 7.3 ± 0.2 at 25°C
Formula may be adjusted and/or supplemented as required to meet performance specifications.

Supplements

NCM4040 Novobiocin

Precaution

Refer to SDS

Preparation

1. Dissolve 43 grams of the medium in 1 Liter of purified water.
2. Autoclave at 121°C for 15 minutes.
3. If required, cool to 45-50°C and add 4 mL of NCM4040-0.5*, Novobiocin Supplement reconstituted using 5mL sterile deionized/RO water.

*Larger vials may be available. Please see appropriate supplement data sheet for availability and preparation instructions.

Quality Control Specifications

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

Prepared Appearance: Prepared medium clarity is clear to lightly hazy with no to light precipitate, and light to medium amber.

Expected Cultural Response: Tryptic Soy Broth Modified with Acid Digest of Casein was inoculated with the test organisms listed on the next page. These organisms were incubated at the appropriate atmosphere and temperature and examined for growth after 18 – 22 hours.

620 Lesher Place • Lansing, MI 48912
800-234-5333 (USA/Canada) • 517-372-9200
foodsafety@neogen.com • foodsafety.neogen.com

Microorganism	Approx. Inoculum (CFU)	Growth
Escherichia coli ATCC® 25922	10 - 300	Growth
Escherichia coli ATCC® 11775	10 - 300	Growth
Escherichia coli ATCC® 35150	10 - 300	Growth
Escherichia coli ATCC® 43888	10 - 300	Growth
Escherichia coli ATCC® 43889	10 - 300	Growth
Escherichia coli ATCC® 43895	10 - 300	Growth
Pseudomonas aeruginosa ATCC® 27853	10 - 300	Markedly Suppressed to Inhibited

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

Refer to appropriate references for specific procedures on the recovery of pathogenic *E. coli*.

Results

Refer to appropriate references for test results on the detection and enumeration of pathogenic *E. coli*.

Expiration

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

Limitations of the Procedure

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

Storage

Store sealed bottle containing the dehydrated medium at 2 - 30°C. 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. USDA. 2013. Food Safety and Inspection Service, Media and Reagents, MLG Appendix 1.08, USDA/FSIS Microbiology Laboratory Guidebook, Washington D.C.
2. U.S. FDA. Center for Food Safety & Applied Nutrition. 2001. Food pathogenic microorganisms and natural toxins handbook. *Escherichia coli* O157:H7. College Park, MD
3. http://www.cdc.gov/ncidod/abmd/diseaseinfo/escherichiacoli_g.htm.
4. Hill, W.E., A. R. Datta, P. Feng, K. A. Lampel, and W. L. Payne. 1998. FDA Bacteriological analytical manual, 8th ed. Identification of Foodborne Bacterial Pathogens by Gene Probes. AOAC International, Gaithersburg, MD.
5. www.fda.gov/Food/ScienceResearch/LaboratoryMethods/BacteriologicalAnalyticalmanualBAM/default.htm.

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