Certificate of Analysis

PREPARED MICROBIOLOGICAL MEDIA



Enterobacteriaceae Enrichment Mossel broth

Catalog Number Product Description Lot Number Date of Manufacture Expiry Date CMS185-9ml-1 Enterobacteriaceae Enrichment Mossel broth, 9ml, 9ml Tube

INTENDED USE

Enterobacteriaceae Enrichment Mossel broth is intended for the selective enrichment of Enterobacteriaceae. The undesired accompanying bacterial flora is inhibited by brilliant green and ox bile. Glucose favors the growth of all Enterobacteriaceae, and the high buffering capacity of the culture medium prevents the formed acid from killing the culture (2). Glucose is the main energy source and supports the rapid growth of all Enterobacteriaceae, while the enzymatic digestion of animal tissue provides nitrogen, vitamins, and amino acids (3).

SUMMARY AND EXPLANATION

EE Broth Mossel Enrichment is prepared according to the formula of Mossel, Visser, and Cornelissen (4). The formula contains dextrose to facilitate the growth of most *Enterobacteriaceae*, thus ensuring the detection of *Salmonella* and other lactose-negative organisms. EE Broth Mossel Enrichment should be used as an enrichment broth, followed by a selective medium; e.g., Violet Red Bile Agar. The enumeration of Enterobacteriaceae is of great concern in monitoring the sanitary condition of food. *Enterobacteriaceae* can be injured in food-processing procedures, which include exposure to low temperatures, sub-marginal heat, drying, radiation, preservatives, or sanitizers (5). Recovery relies on proper results-citation of damaged cells. EE Broth Mossel Enrichment is used to detect and enumerate *Enterobacteriaceae* found per milliliter or per gram of test sample of food when performing the Most Probable Number (MPN) technique with pre-enrichment (6, 7) EE Broth Mossel Enrichment is listed in the USP as one of the recommended media for the isolation of bile-tolerant gram-negative bacteria from nonsterile pharmaceutical products.

PRINCIPLE

Enterobacteriaceae Enrichment Mossel broth is intended for the selective enrichment of *Enterobacteriaceae* from food, animal feed, and other materials.

Peptones provide nitrogen, vitamins, and amino acids. Dextrose is a carbon source. Disodium phosphate and monopotassium phosphate are buffering agents. Brilliant green and oxgall are selective agents.

REAGENTS (FORMULA)

Pancreatic Digest of Gelatin 10.0	g
Glucose5	g
Disodium Hydrogen Phosphate 6.45	g
Potassium Dihydrogen Phosphate 2	g
Ox-bile20	g
Brilliant Green 0.015	g
Deionized Water 1000.0	ml

PROCEDURE

For food samples, refer to appropriate standard references for details on test methods for performing the MPN technique with enrichment using EE Broth Mossel Enrichment.6,7 For pharmaceutical samples, refer to the USP General Chapter for details on the examination of nonsterile products and tests for isolating *Enterobacteriaceae* using EE Broth Mossel Enrichment (1).

EXPECTED RESULTS

Acid production causes the color of EE Broth Mossel Enrichment to become yellow. A negative reaction results in no color change and the medium remains green.

- 1. Suspend 45 g of the powder in 1 L of purified water.
- 2. Heat with frequent agitation until dissolved. DO NOT OVERHEAT. Media is heat-sensitive.
- 3. Dispense into tubes or bottles as required.
- 4. Heat at 100°C in a water bath or flowing steam for 30 minutes. DO NOT AUTOCLAVE.
- 5. Test samples of the finished product for performance using stable, typical control cultures.

QUALITY CONTROL

QA Testing:	Result:	Expected:
Characteristics	Pass	Pass
Sterility	Pass	Pass
Performance	Pass	Pass

pH 7.2 ± 0.2 @ 25°C Appearance Light Green, Clear Storage Condition Refrigerate, 2-8°C

Sterility Method DO NOT AUTOCLAVE

Sterility Test Pass

(Absence of growth following 72 hours at 30 - 37°C)

Analysis Note

Growth promotion test in accordance with the current version of DIN EN ISO 11133.

Mixed inoculum E. coli resp. Salmonella and E.faecalis.

Additionally monoculture of *E.faecalis*

Confirmation on VRBD agar resp. Tryptic soy agar

Inoculum on reference medium (Escherichia coli ATCC* 8739 $(WDCM\ 00012)): \le 100$ Inoculum on reference medium (Escherichia coli ATCC 25922 (WDCM 00013)): < 100 Inoculum on reference medium (Salmonella typhimurium ATCC 14028 (WDCM 00031)): ≤ 100 Inoculum on reference medium (Salmonella enteritidis ATCC 13076 (WDCM 00030)): ≤ 100 Inoculum on reference medium (*Enterococcus faecalis* ATCC 19433 (WDCM 00009)): \geq 1000 Inoculum on reference medium (*Enterococcus faecalis* ATCC 29212 (WDCM 00087)): \geq 1000 Growth on VRBD-Agar(Escherichia coli ATCC 8739 (WDCM 00012)): >10 colonies Growth on VRBD-Agar(Escherichia coli ATCC 25922 (WDCM 00013)): >10 colonies Growth on VRBD-Agar(Salmonella typhimurium ATCC 14028 (WDCM 00031)): >10 colonies Growth on VRBD-Agar(Salmonella enteritidis ATCC 13076 (WDCM 00030)): >10 colonies Growth on Tryptic soy agar (Enterococcus faecalis ATCC 19433 (WDCM 00009)): total inhibition Growth on Tryptic soy agar (Enterococcus faecalis ATCC 29212 (WDCM 00087)): total inhibition Colonies (Escherichia coli ATCC 8739 (WDCM 00012)): pink to red colonies with or without precipitation halo Colonies (Escherichia coli ATCC 25922 (WDCM 00013)): pink to red colonies with or without precipitation halo Colonies (Salmonella typhimurium ATCC 14028 (WDCM 00031)): pink to red colonies with or without precipitation halo

Colonies (Salmonella enteritidis ATCC 13076 (WDCM 00030)): pink to red colonies with or without precipitation halo

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American Public H	ealth Associa	ition, Washingt	on, D.C						

Released By: Mehdi Kargar

Date:

All our prepared media products are manufactured at our site in RCFFN, University of Manitoba, and tested both at our site and by the Department of Microbiology, University of Manitoba.

The generation of this certificate confirms all sterilization and performance criteria have been achieved.

NOTE: Expiry Date only valid if packs are stored unopened at an Ambient Room Temperature not exceeding 25°C.



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