Technical Information

MUG EC Broth

Product Code: DM 2042

Application: - MUG EC Broth is recommended for the detection of *Escherichia coli* in water and food samples by a fluorogenic method.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	20.000
Lactose	5.000
Bile salts mixture	1.500
Dipotassium phosphate	4.000
Monopotassium phosphate	1.500
Sodium chloride	5.000
4-Methylumbelliferyl ß-D-Glucuronide (MUG)	0.050
Final pH (at 25°C)	6.9±0.2

**Formula adjusted, standardized to suit performance parameters

Principle & Interpretation

MUG EC Broth is also used by APHA for the analysis of drinking water, surface and ground water and waste-water for the presence of *E.coli* (2). MUG permits rapid detection of *E. coli* when medium is observed for fluorescence using UV Light (3, 4). *Escherichia coli* is a member of faecal coliform group of bacteria. It is a member of the indigenous faecal flora of warm- blooded animals. *E.coli* is considered a specific indicator of faecal contamination and the possible presence of enteric pathogens. EC Broth was devised by Hajna and Perry (1) and further modified by addition of the fluorogenic compound MUG. MUG also detects anaerogenic strains which may not be detected in conventional procedure (3). MUG is hydrolyzed by the enzyme #-glucuronidase possessed by *E.coli* to yield a fluorescent end product 4-Methylumbelliferone.

Casein enzymic hydrolysate supplies essential nutrients. Lactose acts as fermentable carbohydrate. Sodium chloride helps to maintain osmotic equilibrium. The medium has a strong buffering system to control the pH in the presence of fermentative action. The bile salts inhibit gram-positive bacteria especially *Bacillus* species and faecal Streptococci. Mostly beta-glucuronidase activity occurs within 4 hours but some weak beta- glucuronidase-positive strains require overnight incubation (2). The fermentation of lactose by lactose fermentors leads to acidification of the medium, resulting in drop of pH. Adjustment of pH of cultures by sodium hydroxide enhanced fluorescence as observed by Maddocks and Greenman (5).Similarly Freir and Hartman (6) noticed that exposure of tubes to ammonia fumes enhanced fluorescence.

Large number of *Proteus vulgaris* if present, may suppress gas production of *E.coli*, however fluorescence permits detection of *E.coli* in pure or mixed cultures within 4 to 24 hours.

Inoculate the test water sample into PA Broth (DM 2186) and Lauryl Sulphate Broth (DM 1080). After an incubation at 35-37°C for 18-24 hours, all presumptive tubes showing growth, gas or acidity is further tested using MUG EC Broth (DM 2042). After an incubation at 35-37°C for 4-24 hours, the presence of a bright blue fluorescence is considered as a positive response for *E. coli.*

Methodology

Suspend 37.05 grams of dehydrated powder media in 1000 ml distilled water. Mix thoroughly & heat, if necessary to dissolve the medium completely. Dispense in tubes containing inverted Durham's tubes. Sterilize by autoclaving at 15 lbs pressure (121°C) for 12-15 minutes.

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Colour and Clarity
Yellow coloured clear solution without any precipitate

Reaction
Reaction of 3.7% w/v aqueous solution at 25°C. pH : 6.9±0.2

pH Range
6.70-7.10

Cultural Response
DM 2042: Cultural characteristics observed after an incubation at 35 - 37°C for 4 - 24 hours.

Organism	Inoculum (CFU)	Growth	Fluorescence (under uv) (at 366 nm)
Escherichia coli ATCC 25922	50-100	luxuriant	positive, throughout the tube
Enterobacter aerogenes ATCC 13048	50-100	luxuriant	negative
Staphylococcus aureus ATCC 25923	>=10^3	inhibited	-
Salmonella Typhi ATCC 6539	50-100	good	negative
Shigella flexneri ATCC 12022	50-100	good	Negative

Storage and Shelf Life

Dried Media: Store below 30°C in tighty closed container and prepared medium below 2-8°C. Use before expiry period on the label.
Prepared Media: 2-8° in sealable plastic bags for 2-5 days.

Further Reading

1. Hajna A. A. and Perry C. A., 1943, Am. J. Public Health, 33:550.
2. Feng P. C. S. and Hartman P. A. S., 1982, Appl. Environ. Microbiol., 43:132.
3. Robinson B. J., 1984, Appl. Environ. Microbiol., 48:285.
4. Greenberg A. E., Trussell R. R. and Clesceri L. S., (Eds.), 1988, Standard Methods for the Examination of Water andWastewater, 20th Ed., APHA, Washington, D.C.
5. Maddocks J. L. and Greenan M. J. (1975) J. Clin. Pathol. 28. 686-687.
6. Freir T. A. and Hartman P. A. (1987) Appl. Env. Microbiol. 53. 1246-1250.

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