Mineral Modified Glutamate Agar Base (Twin Pack) M643I

Intended Use

Recommended for the enumeration of *Escherichia coli* from meat and meat products. The composition and performance criteria of this medium are as per the specifications laid down in ISO 6391:1997, ISO 11866-1:2005(E) IDF 170-1:2005(E) and IS 15463: 2004.

Composition**

ISO 6391:1997 ISO 11866-1:2005(E) IDF 170-1:2005(E) and IS 15463: 2004. Specification-Mineral Modified Glutamate Agar

Ingredients	g / L
Sodium glutamate	6,35 g
Lactose	10,0 g
Sodium formate	0,25 g
L(-)-cystine	0,02 g
L(-)-aspartic acid	0,024 g
L(+)-arginine	0,02 g
Thiamine	0,001 g
Nicotinic acid	0,001 g
Pantothenic acid	0,001 g
Magnesium sulfate (MgSO4.7H2O)	0,100 g
Ammonium iron(II) citrate	0,010 g
Calcium chloride (CaCl2.2H2O)	0,010 g
Dipotassium hydrogen phosphate	0,90 g
Ammonium chloride	2,5 g
Agar	12 g to 18g
Final pH (at 25°C)	6.7±0.2

**Formula adjusted, standardized to suit performance parameters

M643I- Mineral Modified Glutamate Agar

Ingredients	g / L
Part A	-
Lactose	10.000
Dipotassium hydrogen phosphate	0.900
Sodium formate	0.250
L-Cystine	0.020
L-Aspartic acid	0.024
L-Arginine	0.020
Thiamine	0.001
Nicotinic acid (Niacin)	0.001
Pantothenic acid	0.001
Ferric ammonium citrate	0.010
Calcium chloride dihydrate	0.010
Magnesium sulphate heptahydrate	0.100
Agar	15.000
Part B	-
Sodium glutamate	6.350
Final pH (at 25°C)	6.7±0.2

Directions

Suspend 26.29 grams (the equivalent weight of dehydrated medium per litre)of Part A and 6.35 grams of Part B in 1000 ml purified / distilled water containing 2.5 grams ammonium chloride. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 10lbs (115°C) for 10 minutes. Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Folpmers (1) described a glutamic acid based chemically defined medium for the enumeration of coliform bacteria from water and wastewater. This glutamate-containing medium was later modified by Gray (2), by the addition of lactose, which gave less false positive results when compared to MacConkey Broth.

Mineral Modified Glutamate Agar Base is recommended for the enumeration of *Escherichia coli* from meat and meat products by colony count at 44°C by membrane filtration technique(3). Anderson and Baird-Parker (4) described a direct plate method for the rapid enumeration of *E. coli* in foods and this method was modified by a resuscitation procedure using Mineral Modified Glutamate Agar with successful recovery of damaged cells from frozen, dried, heat processed or low pH foods. Mineral Modified Glutamate Agar Base (Twin Pack)is also recommended by ISO Committee(5)

This media contain a variety of nutrients including salts, amino acids and vitamins. Lactose is the fermentable carbohydrate. Because of the nutrients, this media is superior for enumerating coliforms in water and wastewater as it satisfies most of the nutritional requirements of coliforms.

Type of specimen

Food samples: meat and meat products

Specimen Collection and Handling:

For food samples, follow appropriate techniques for sample collection and processing as per guidelines (6).
After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/ face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets

Limitations :

1. Due to variable nutritional requirements, some strains show poor growth on this medium.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Part A : Off-white to light yellow homogeneous free flowing powder Part B : Colourless to white needles

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of the medium (2.634% w/v Part A + 0.635% w/v Part B + 0.25% ammonium chloride) is at 25°C. pH : 6.7±0.2

pH

6.50-6.90

Cultural Response

Cultural characteristics observed after an incubation at 44°C for 24 hours.

Organism	Inoculum (CFU)	Growth	Recovery
Escherichia coli ATCC 25922 (00013*)	50-100	luxuriant	>=50%

Key : *Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (7,8).

Reference

1. Folpmers T., 1948, Ant. V. Leeuwenhoek, J. Microbiol. Serol., 14:58.
2. Gray R.D., 1959, J. Hyg. Camb., 57:249.
3. Meat and meat products -- Enumeration of *Escherichia coli* International Organization for Standardization (ISO), 1988, Draft, ISO/DIS 6391.
4. Anderson J. M., and Baird Parker A. C., 1975, J. Appl. Bacteriol., 39:111.
5. Milk and milk products — Enumeration of presumptive *Escherichia coli* —Part 1:Most probable number technique using 4-methylumbelliferyl-ß-D-glucuronide (MUG) International Organization for Standardization (ISO),ISO 11866-1:2005(E) IDF 170-1:2005(E)
6. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
7. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
8. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

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