Fluid Thioglycollate Medium

M009B

Fluid Thioglycollate Medium is used for sterility testing of biologicals and for cultivation of aerobes, anaerobes and microaerophiles as per British Pharmacopoeia.

Composition**

Ingredients	Gms / Litre
Tryptone#	15.000
Yeast extract	5.000
Glucose monohydrate	5.500
Sodium chloride	2.500
L-Cystine	0.500
Sodium thioglycollate	0.500
Resazurin sodium	0.001
Agar	0.750
pH after sterilization (at 25°C)	7.1±0.2

**Formula adjusted, standardized to suit performance parameters

- Equivalent to Pancreatic digest of casein

Directions

Suspend 29.25 grams (the equivalent weight of dehydrated medium per litre) in 1000 ml Water R/purified/distilled water. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 25°C and store in a cool dark place preferably below 25°C.

Note : If more than the upper one-third of the medium has acquired a pink colour, the medium may be restored once by heating in a water bath or in free flowing steam until the pink colour disappears.

Principle And Interpretation

Brewer (1) formulated Fluid Thioglycollate Medium for rapid cultivation of aerobes as well as anaerobes including microaerphiles by adding a reducing agent and small amount of agar. The British Pharmacopoeia (2) USP (3), EP (4) and AOAC (5) have recommended the media for sterility testing of antibiotics, biologicals and foods and for determining the phenol coefficient and sporicidal effect of disinfectants. However, it is intended for the examination of clear liquid or water-soluble materials.

Tryptone and yeast extract serves as a source of nitrogen, carbon, long chain amino acids. vitamins and other essential growth nutrients. Glucose monohydrate is the fermentable carbohydrate and energy source. L-cystine is the amino acid necessary for bacterial multiplication. Sodium thioglycollate and L-cystine act as a reducing agent lowering the oxidation-reduction potential by removal of oxygen. This condition helps to prevent the accumulation of peroxides which is toxic in nature. The SH group of cystine also neutralizes the antibacterial effect of mercurial preservatives and other heavy metal compounds which exert a bacteriostatic effect in the materials under examination. Any increase in the oxygen content is indicated by a colour change of redox indicator, resazurin to red (6,7,8). The small amount of agar helps in maintaining low redox potential for stabilizing the medium (9).

In sterility checking, it is recommended to dilute the sample containing preservatives, with this broth to reduce the toxicity and enhance the growth of contaminants, if any.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Light straw coloured clear to slightly opalescent solution with upper 10% or less medium pink on standing.

Reaction

After sterilization,reaction of 2.92% w/v aqueous solution. pH : 7.1±0.2

pH
6.90-7.30

Growth Promotion Test
As per British Pharmacopoeia

Growth promoting properties
Clearly visible growth of microorganism comparable to that previously obtained with previously tested and approved lot of medium occurs at the specified temperature for not more than the shortest period of time specified inoculating <=100 cfu(at 30-35°C for <=3 days.

Stability test
Light yellow coloured clear solution without any precipitation sedimentation at room temperature for 7 days

Cultural Response
M009B: Cultural characteristics observed after an incubation at 30-35°C for not more than 3 days.

Organism	Inoculum (CFU)	Growth
Clostridium sporogenes ATCC 19404	50 -100	luxuriant
Clostridium sporogenes ATCC 11437	50 -100	luxuriant
Clostridium perfringens ATCC 13124	50 -100	luxuriant
Bacteroides fragilis ATCC 23745	50 -100	luxuriant
Bacteroides vulgatus ATCC 8482	50 -100	luxuriant
Staphylococcus aureus ATCC25923	50 -100	luxuriant
Staphylococcus aureus ATCC 6538	50 -100	luxuriant
Pseudomonas aeruginosa ATCC 27853	50 -100	luxuriant
Pseudomonas aeruginosa ATCC 9027	50 -100	luxuriant
Micrococcus luteus ATCC 9341	50 -100	luxuriant
Escherichia coli ATCC 25922	50 -100	luxuriant
Escherichia coli ATCC 8739	50 -100	luxuriant
Escherichia coli NCTC 9002	50 -100	luxuriant
Salmonella Typhimurium ATCC 14028	50 -100	luxuriant
Salmonella Abony NCTC 6017	50 -100	luxuriant
Bacillus subtilis ATCC 6633	50 -100	luxuriant

Storage and Shelf Life
Store below 30°C in tightly closed container and the prepared medium at room temperature and away from light. Use before expiry date on the label.

Reference
1.Brewer, 1940, J. Am. Med. Assoc., 115:598.
2.British Pharmacopoeia, 2014, The Stationery office British Pharmacopoeia.
3.U.S. Pharmacopoeia, 2014, United States Pharmacopoeia Convention, Inc., Rockville, MD.
4.European Pharmacopoeia, 2014, European Department, for the Quality of Medicines.
5. Williams. (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th ed., AOAC, Washington, D.C.
6.Marshall, Gunnison and Luxen, 1940, Proc. Soc. Exp. Biol. Med., 43:672.
7.Nungester, Hood and Warren, 1943, Proc. Soc. Exp. Biol. Med., 52:287.

8.Portwood, 1944, J. Bact., 48:255.
9 MacFaddin J.F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of ,,Medical Bacteria, Vol.1, Williams and Wilkins, Baltimore.

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