

Technical Data

Alternative Thioglycollate Medium (NIH Thioglycollate Broth) M010 (Thioglycollate Broth, Alternative)

Intended Use:

Alternative Thioglycollate Medium is recommended for sterility testing of turbid or viscous biological products.

Composition**

Ingredients	Gms / Litre
Tryptone	15.000
Yeast extract	5.000
Dextrose (Glucose)	5.500
Sodium chloride	2.500
L-Cystine	0.500
Sodium thioglycollate	0.500
Final pH (at 25°C)	7.1±0.2

^{**}Formula adjusted, standardized to suit performance parameters

Directions

Suspend 29.0 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Mix well and dispense into sterile tubes or flasks as desired. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Note: It is preferable to use freshly prepared medium, alternatively it should be boiled and cooled just once prior to use as on reheating, toxic oxygen radicles are formed.

Principle And Interpretation

Alternative Thioglycollate Medium is formulated as described in the N.I.H. memorandum (5). It is used for the sterility testing of certain biological products which are turbid or viscous and cant be tested using Fluid Thioglycollate Medium (M009). Both the media have similar composition, except agar and resazurin that are not included in Alternative Thioglycollate Medium. This deletion makes it suitable for sterility testing of viscous products.

Tryptone serves as a source of nitrogen and carbon compounds, long chain amino acids and other essential nutrients. Yeast extract serve as source of essential nutrients to the contaminants, if present. Dextrose serves as the energy source. Sodium chloride maintains the osmotic equilibrium of the medium whereas L-cystine, an amino acid, also serves as source of essential growth factors. Sodium thioglycollate and L-cystine lower the oxidation-reduction potential of the medium by removing oxygen to maintain a low Eh. Sodium thioglycollate also helps to neutralize the toxic effects of mercurial preservatives (6,7).

Type of specimen

Clinical: wound swabs, skin swabs or scrapings, tooth tartar etc. Pharmaceutical: Sterility testing of viscous products.

Specimen Collection and Handling

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (1,2).

For pharmaceutical products, follow appropriate techniques for sample processing in case of viscous materials as mentioned under sterility. (8)

Warning and Precautions

In Vitro diagnostic use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

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Limitations

- 1. The tubes should not be reheated as frequent boiling leads to development of toxic products.
- 2. Prior to use the medium should be boiled once to remove the absorbed oxygen.
- 3.Before inoculation, the tubes should be brought to room temperature
- 4. The medium should not be used in fermentation process as medium contains yeast extract which is high in carbohydrate content.

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

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

yellow coloured clear solution without any precipitate.

Reaction

Reaction of 2.9% w/v aqueous solution at 25°C. pH: 7.1±0.2

pН

6.90-7.30

Cultural Response

Cultural characteristics observed after an incubation at 30-35°C for not more than 3 days.(*Incubated anaerobically).

Cultural Response

M010: Complies

Organism	Inoculum (CFU)	Growth
Cultural Response		
Clostridium sporogenes ATCC 19404 (00008)	50 -100	luxuriant
*Clostridium sporogenes ATCC 11437	50 -100	luxuriant
*Clostridium sporogenes NBRC 14293	50 -100	luxuriant
Clostridium perfringens ATCC 13124 (00007)	50 -100	luxuriant
Staphylococcus aureus subsp. aureus ATCC 25923 (00034*)	50 -100	luxuriant
Staphylococcus aureus subsp. aureus ATCC 6538 (00032*)	50 -100	luxuriant
Pseudomonas aeruginosa ATCC 27853 (00025*)	50 -100	luxuriant
Pseudomonas aeruginosa ATCC 9027 (00026*)	50 -100	luxuriant
Escherichia coli ATCC 25922 (00013*)	50 -100	luxuriant
Escherichia coli ATCC 8739 (00012*)	9 50 -100	luxuriant
Escherichia coli NCTC 9002	2 50 -100	luxuriant
Salmonella Abony NCTC 6017 (00029*)	50 -100	luxuriant
Salmonella Typhimurium ATCC 14028 (00031*)	50 -100	luxuriant
*Bacteroides fragilis ATCC 23745	50 -100	luxuriant
*Bacteroides vulgatus ATCC 8482	C 50 -100	luxuriant
Key: *Corresponding WDCM numbers.		

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Storage and Shelf Life

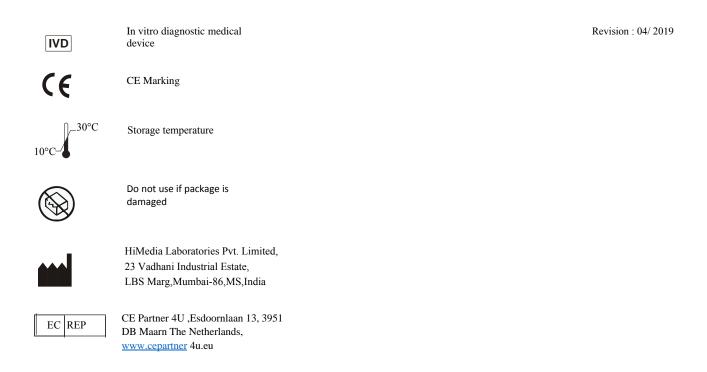
Store between 10-30°C in tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle inorder to prevent lump formation due to the hygroscopic nature of the product. Improper storage 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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (1,2).

Reference

- 1.Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
- 2. 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.
- 3. Lapage S., Shelton J. and Mitchell T., 1970, Methods in Microbiology', Norris J. and Ribbons D., (Eds.), Vol. 3A, Academic Press, London.
- 4. MacFaddin J. F., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd Ed., Lippincott, Williams and Wilkins, Baltimore.
- 5. N.I.H. Memorandum, 1955: Culture Media for Sterility Tests, 4th Revision.
- 6. Nungester, Hood and Warren, 1943, Proc. Soc. Exp. Biol. Med., 52: 287
- 7. Portwood, 1944, J. Bacteriol., 48: 255
- 8. The United States Pharmacopoeia, 2019, The United States Pharmacopoeial Convention. Rockville, MD.



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