

**Fluid Thioglycollate Medium****84681.0500****Also known as**

Thioglycollate Medium, Fluid

**Intended use**

Media for sterility testing of biologicals and for cultivation of aerobes, anaerobes and microaerophiles.

**Formula\*\* - Composition in g/L**

<b>Ingredients</b>	<b>g/L</b>
Pancreatic digest of casein	15.000
Yeast extract	5.000
Dextrose	5.500
Sodium chloride	2.500
L-Cystine	0.500
Sodium thioglycollate	0.500
Resazurin sodium	0.001
Agar	0.750
Final pH ( at 25°C)	7.1±0.2

\*\*Formula adjusted, standardised to suit performance parameters

**Instructions for preparation**

Suspend 29.75 grams in 1000 ml purified / distilled water. Heat to boiling to dissolve the medium completely. Sterilise 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 of the method and general information**

Brewer (1) formulated Fluid Thioglycollate Medium for rapid cultivation of aerobes as well as anaerobes including microaerophiles by adding a reducing agent and small amount of agar. The USP (2), BP(3), EP(4) have and AOAC (5) 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. Fluid Thioglycollate Medium is also routinely used to check the sterility of stored blood in blood banks(10).

Dextrose, pancreatic digest of casein, yeast extract, L-cystine provide the growth factors necessary for bacterial multiplication. L-cystine and sodium thioglycollate allows *Clostridium* to grow in this medium even under aerobic conditions (11). Also the small amount of agar used in the medium favors the growth of aerobes as well as anaerobes in the medium, even if sodium thioglycollate is deleted from the medium(1). Sodium thioglycollate act as a reducing agent and neutralizes the toxic effects of mercurial preservatives and peroxides formed in the medium, thereby promoting anaerobiosis, and making the medium suitable to test materials containing heavy metals. (9,10). 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 stabilising the medium (9).

### Instruction for use

1. This medium can be dispensed in tubes or bottles for use.
2. Dispense 15 ml of media in tubes of 20 x 150 mm size.
3. Inoculate sample or its dilutions carefully at the bottom of the tubes.
4. To create strict anaerobic conditions, 1 ml of sterile agar or sterile paraffin solution should be overlaid.
5. This medium is also used in pharmaceuticals for sterility testing. The procedures specified in Pharmacopoeia, USP should be followed for testing the sample and test result interpretation.

### Limitations

1. This medium supports growth of aerobes, anaerobes as well as microaerophiles and facultative anaerobes. Some anaerobes may get inhibited by metabolic end products or acids formed due to rapidly growing facultative anaerobes. For strict isolation of anaerobes, growth obtained in broth cultures must be subcultured on solid media to obtain pure cultures.
2. For optimal recovery of strict aerobes, a second liquid enrichment medium such as Brain Heart Infusion Broth (84626.0500) or Soyabean Casein Digest Medium (84675.0500) should be used.
3. FTM is also used for testing clinical and nonclinical specimens. For optimal recovery of fastidious anaerobes, e.g., *Prevotella* spp., the medium should be supplemented with hemin and vitamin K1.

### 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

Reaction of 2.97% w/v aqueous solution at 25°C. pH : 7.1±0.2

#### pH

6.90-7.30

### Cultural Response

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

Cultural Response

Organism	Inoculum (CFU)	Growth
<i>Clostridium sporogenes</i> ATCC 19404	50 -100	good
<i>Clostridium sporogenes</i> ATCC 11437	50 -100	good
<i>Clostridium sporogenes</i> NBRC 14293	50 -100	good
<i>Clostridium perfringens</i> ATCC 13124	50 -100	good
<i>Bacteroides fragilis</i> ATCC 23745	50 -100	good
<i>Bacteroides vulgatus</i> ATCC 8482	50 -100	good
<i>Staphylococcus aureus</i> ATCC25923	50 -100	good
<i>Staphylococcus aureus</i>	50 -100	good

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ATCC 6538		
<i>Pseudomonas aeruginosa</i>	50 -100	good
ATCC 27853		
<i>Pseudomonas aeruginosa</i>	50 -100	good
ATCC 9027		
<i>Micrococcus luteus</i> ATCC 9341	50 -100	good
<i>Streptococcus pneumoniae</i> ATCC 6305	50 -100	good
<i>Escherichia coli</i> ATCC 25922	50 -100	good
<i>Escherichia coli</i> ATCC 8739	50 -100	good
<i>Escherichia coli</i> NCTC 9002	50 -100	good
<i>Salmonella</i> Typhimurium ATCC 14028	50 -100	good
<i>Salmonella</i> Abony NCTC 6017	50 -100	good
<i>Bacillus subtilis</i> ATCC 6633	50 -100	good

#### Reference

1. Brewer, 1940, J. Am. Med. Assoc., 115:598.
2. The United States Pharmacopoeia, 2009, The United States Pharmacopoeial Convention, Rockville, MD.
3. British Pharmacopoeia, 2009, The Stationery office British Pharmacopoeia
4. European Pharmacopoeia, 2009, European Dept. for the quality of Medicines.
5. Williams H., (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.
10. Federal Register, 1992, Fed. Regist., 21:640.2.17.
11. Quastel and Stephenson, 1926, J. Biochem., 20:1125.

#### Storage conditions

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry period on the label.

#### Ordering information

84681.0500	Fluid Thioglycollate Medium	Bottle of 500g
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