PRODUCT SPECIFICATION SHEET

Thioglycollate Medium w/o Dextrose (DM496)

Intended Use
Thioglycollate Medium w/o Dextrose (DM496) is recommended for cultivation of aerobes, microaerophiles, anaerobes and for carbohydrate fermentation studies

Product Summary and Explanation
Quastel and Stephenson[1] noticed that the presence of a small amount of a compound containing an –SH group (cysteine, thioglycollic acid, glutathione) allowed "aerobic" growth of *Clostridium sporogenes* in tryptic digest broth.
Falk, Bucca and Simmons[2] brought out the advantages of using small quantities of agar (0.06-0.25%) in detecting contaminants during sterility testing of biologicals. Brewer[3] demonstrated the value of combining a small amount of agar and a reducing substance. Brewer's experiments revealed that anaerobes grew equally well, in a liquid medium containing 0.05% agar, irrespective of presence or absence of sodium thioglycollate. Marshall, Gunnish and Luxen[4] reported satisfactory cultivation of anaerobes in Brewer's Thioglycollate Medium in the presence of a mercurial preservative. Neutralization of the bacteriostatic effect of mercurial compounds by sodium thioglycollate was confirmed by Nungester, Hood and Warren[5] and Portwood.[6] Vera[7] introduced incorporation of casein peptone. Malin and Finn[8] reported the commonly used medium containing thioglycollate is inhibitory to some organisms in the presence of a carbohydrate.
Fluid Thioglycollate Medium is recommended in the FDA *Bacteriological Analytical Manual* (BAM)[9] and the *Official Methods of Analysis of AOAC International* [10] for the examination of food, and for determining the phenol coefficient and sporicidal effects of disinfectants. Fluid Thioglycollate Medium is also specified for sterility checks on banked blood.[11] It is one of the media recommended in the *USP* for use in sterility testing of articles purporting to be sterile; these formulations meet the requirements of the *USP* growth promotion test.[12]
Thioglycollate Medium without Dextrose is the modification of original Thioglycollate medium[3, 13] used for the fermentation study of anaerobes and for enhancement of sporulation. Omission of dextrose facilitates it to be used in fermentation studies with the addition of desired carbohydrate. Some Clostridia remain viable for a longer period and sporulate better in the absence of carbohydrate and thus this medium could be used for sporulations.

Principles of the Procedure
Thioglycollate Medium w/o Dextrose contains casein enzymic hydrolysate, L-cystine and salts which provides essential nutrients like nitrogenous compounds, carbon, sulphur, minerals and amino acids necessary for bacterial multiplication. The reducing action provided by sodium thioglycollate and sodium sulphite binds molecular oxygen, thereby maintaining a low Eh. A small amount of agar is added to retard the absorption of oxygen by reducing convection currents in the medium. Methylene blue is an indicator dye when oxygen content increases slightly; it is indicated by a colour change of redox indicator.

Formula / Liter

Ingredients	Gms / Liter
Casein enzymic hydrolysate	20.00
Sodium chloride	2.50
Dipotassium phosphate	1.50
Sodium thioglycollate	0.60
L-Cystine	0.40
Sodium sulphite	0.20
Methylene blue	0.002
Agar	0.50

PRODUCT SPECIFICATION SHEET

Final pH: 7.2 ± 0.2 at 25°C
Formula may be adjusted and/or supplemented as required to meet performance specifications

Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.
3. If more than the upper one-third of the medium has acquired a green colour, the medium may be restored once by heating in a water bath or in free flowing steam until the green colour disappears.
4. Do not reheat the media more than once; continued reheating gives rise to toxicity.

Directions

1. Suspend 25.7 grams of the medium in one liter of distilled water.
2. Heat to boiling, to dissolve the medium completely.
3. If the medium is to be used for fermentation studies or for diagnostic work adds 0.5 to 1% carbohydrate of choice.
4. Dispense and autoclave at 121°C, 15 psi pressure, for 15 minutes / validated cycle.
5. Alternatively, sterile carbohydrate solutions may be added to the broth after sterilization.
6. The prepared medium should be stored in the dark at room temperature.

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Light yellow coloured very slightly opalescent viscous solution with upper 10% or less medium green on standing
Reaction of 2.57% Solution	pH : 7.2 ± 0.2 at 25°C
Gel Strength	Not Applicable

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 48 hours (in an appropriate atmosphere).

Sr. No.	Organisms	Results to be achieved	
		Inoculum (CFU)	Growth
1.	*Bacillus subtilis ATCC 6633*	50-100	good
2.	*Bacteroides vulgatus ATCC 8482*	50-100	fair
3.	*Candida albicans ATCC 10231*	50-100	good
4.	*Clostridium sporogenes ATCC 11437*	50-100	good-luxuriant
5.	*Micrococcus luteus ATCC 10240*	50-100	good
6.	*Neisseria meningitidis ATCC 13090*	50-100	good
7.	*Streptococcus pyogenes ATCC 19615*	50-100	good-luxuriant

The organisms listed are the minimum that should be used for quality control testing.

Test Procedure

Refer to appropriate references for standard test procedures.

Results

1. After incubation, growth is evidenced by the presence of turbidity compared to an uninoculated control.

2. Strict aerobes tend to grow in a thin layer at the surface of the broth; obligate anaerobes will grow only in that portion of the broth below the upper oxidized layer.

Storage

Store the sealed bottle containing the dehydrated medium at 10- 30°C. Once opened and recapped, place container in a low humidity environment at the same storage temperature. Protect from moisture and light.

Expiration

Refer to the expiration date stamped on the container. The dehydrated medium should be discarded if not free flowing, or if the appearance has changed from the original color. Expiry applies to medium in its intact container when stored as directed.

Limitations of the Procedure

1. Anaerobes can be overgrown by more rapidly growing facultative organisms.
2. If plating medium reveals no growth examine and Gram stain broth.
3. Never rely on broth cultures exclusively for isolation of anaerobes. Some anaerobes may be inhibited by metabolic products or acids produced from more rapidly growing facultative anaerobes.
4. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : Thioglycollate Medium w/o Dextrose
Product Code : DM496
Available Pack sizes : 500gm

References

1. Quastel and Stephenson. 1926. J. Biochem. *20*:1125.
2. Falk, Bucca and Simmons. 1939. J. Bacteriol. *37*:121.
3. Brewer. 1940. JAMA *115*:598.
4. Marshall, Ginnish and Luxen. 1940. Proc. Soc. Exp. Biol. Med. *43*:672.
5. Nungester, Hood and Warren. 1943. Proc. Soc. Exp. Biol. Med. 52:287.
6. Portwood. 1944. J. Bacteriol. *48*:255.
7. Vera. 1944. J. Bacteriol. *47*:59.
8. Malin and Finn. 1957. J. Bacteriol. *62*:349.
9. U.S. Food and Drug Administration. 2001. Bacteriological analytical manual, online. AOAC International, Gaithersburg, Md.
10. Horwitz (ed.). 2007. Official methods of analysis of AOAC International, 18th ed., online. AOAC International, Gaithersburg, Md.
11. Federal Register. 1992. Fed. Regist. *21*:640.2.17.
12. United States Pharmacopeial Convention, Inc. 2008. The United States pharmacopeia 31/The national formulary 26, Supp. 1, 8-1-08, online. United States Pharmacopeial Convention, Inc., Rockville, Md.
13. Brewer J. H., 1940, J. Bacteriol., 39:10.

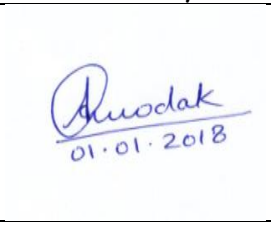
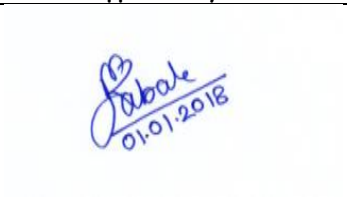
Further Information

For further information please contact your local MICROMASTER Representative.

PRODUCT SPECIFICATION SHEET

MICROMASTER LABORATORIES PRIVATE LIMITED
Unit 38/39, Kalpataru Industrial Estate,
Off G.B. Road, Near 'R-Mall' , Thane (W) – 400607. M.S. INDIA.
Ph: +91-22-25895505, 4760, 4681. Cell: 9320126789.
Email: micromaster@micromasterlab.com
sales@micromasterlab.com

DM496PSS,QAD/FR/024,Rev.00/01.01.2018

Prepared By	Checked By	Approved By
		
Microbiologist	**Head Quality Control**	**Head Quality Assurance**

Disclaimer :

All Products conform exclusively to the information contained in this and other related Micromaster Publications. Users must ensure that the product(s) is appropriate for their application, prior to use. The information published in this publication is based on research and development work carried out in our laboratory and is to the best of our knowledge true and accurate. Micromaster Laboratories Pvt Ltd reserves the right to make changes to specifications and information related to the products at any time. Products are intended for laboratory, diagnostic, research or further manufacturing use only and not for human or animal or therapeutic use, unless otherwise specified. Statements included herein should not be considered as a warranty of any kind, expressed or implied, and no liability is accepted for infringement of any patents.