PRODUCT SPECIFICATION SHEET

M-Staphylococcus Broth (DM541)

Intended Use

M-Staphylococcus Broth (DM541) is recommended for detection and isolation of Staphylococci by MF technique.

Product Summary and Explanation

The presence of staphylococci, along with other organisms, are indicators of recreational water quality. [1] *Pseudomonas*, *Streptococcus* , and *Staphylococcus* are normal skin flora that are likely to be shed and are indicators of health risk. [2]These organisms account for a large percentage of swimming pool-associated illness. [1] Staphylococci are widespread in nature, though they are mainly found living on the skin, skin glands, and mucous membranes of mammals and birds.[3] m Staphylococcus Broth is patterned after the formula of Staphylococcus Medium 110 that Chapman created while developing a selective medium for staphylococci. [4] For the multiple-tube procedure to monitor swimming pool water for *S. aureus* , sodium azide is added to m Staphylococcus Broth. [1]m Staphylococcus Broth contains peptone to provide the nitrogen, amino acids, and minerals. Yeast extract provides the vitamin source. Lactose and mannitol are carbohydrates for bacterial growth. Dipotassium phosphate is the buffering agent and the high concentration of sodium chloride causes the medium to be selective for staphylococci.

Principles of the Procedure

Staphylococci are gram-positive cocci residing on the skin and mucous membrane of humans and other organisms. M-Staphylococcus Broth is used for detection and isolation of Staphylococci by membrane filter technique. This broth is especially used for isolating pathogenic and enterotoxigenic Staphylococci and has similar composition as Staphylococcus Agar No. 110 except agar and gelatin (1). Casein enzymic hydrolysate and yeast extract supply essential growth factors such as nitrogen, carbon, sulphur, vitamins and trace ingredients. The 7.5% concentration of sodium chloride results in partial or complete inhibition of bacteria except Staphylococci. Mannitol and lactose are utilized as energy sources. Inoculate the tubes of M-Staphylococcal Broth and incubate at 35 ± 2°C for 24 hours. Streak from positive tubes (turbid growth) on plates of Lipovitellin Salt Mannitol Agar Base (M627) and incubate at 35-37°C for 48 hours. Opaque, yellow zones around the colonies are positive evidence of lipovitellin- lipase activity and mannitol fermentation (2). Alternatively around 2 ml of M-Staphylococcus Broth is used to saturate sterile absorbent cotton pads. Membrane filters used for filtration are aseptically placed on these saturated cotton pads. Following an incubation at 35-37°C for 18-48 hours, observe membrane for growth and pigment production. Mannitol fermentation can be visualized as yellow colouration by addition of a few drops of bromothymol blue to the areas from where colonies have been removed.

Formula / Liter

Ingredients	Gms / Liter
Casein enzymic hydrolysate	10.000
Yeast extract	2.500
Lactose	2.000
Dipotassium hydrogen phosphate	5.000
Sodium chloride	75.000
Sodium azide	0.049
Final pH: 7.0 ± 0.2 at 25°C	
Formula may be adjusted and/or supplemented as required to meet performance specifications	

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Precautions

1. For Laboratory Use only.
2. IRRITANT. Irritating to eyes, respiratory system, and skin.

Directions

1.Suspend 104.55 grams in 1000 ml distilled water.
2.Mix thoroughly and heat to boiling for 5 minutes.
3.DO NOT AUTOCLAVE. For 10 ml inocula, use double strength medium.
Warning : Sodium azide has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables

Quality Control Specifications

Dehydrated Appearance	Cream to yellow homogeneous free flowing powder
Prepared Medium	Light amber coloured clear solution without any precipitate
Reaction of 3.42% Solution	pH : 7.0 ± 0.2 at 25°C
Gel Strength	Not Applicable

Expected Cultural Response: Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours .

Sr. No.	Organisms	Results to be achieved	
		Inoculum (CFU)	Growth
1.	*Enterococcus faecalis ATCC 29212*	>=10^3	inhibited
2.	*Escherichia coli ATCC 25922*	>=10^3	inhibited
3.	*Staphylococcus aureus ATCC 25923*	50-100	good-luxuriant
4.	*Staphylococcus epidermidis ATCC 12228*	50-100	good-luxuriant
5.	*Streptococcus pyogenes ATCC 19615*	>=10^3	inhibited

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

Test Procedure

Refer appropriate references for specific test procedures.

Results

Refer appropriate references and test procedures for interpretation of results.

Storage

Store the sealed bottle containing the dehydrated medium at 2 - 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.

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Limitations of the Procedure

1. If the medium is not used on same day, it is advisable to drive off dissolved gases by boiling or steaming in the autoclave and cool without agitation.
2. While reheating prepared media to drive off dissolved gases, do not overheat because this may result in decreased growth.
3. Consult appropriate texts for detailed information and recommended procedures.

Packaging

Product Name : M-Staphylococcus Broth
Product Code : DM541
Available Pack sizes : 500gm

References

1.MacFaddin J. F., 1985, Media for Isolation-Cultivation-Maintenance-of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.

2. Eaton A. D., Clesceri L. S. and Greenberg A. E., (Eds.), 1995, Standard Methods for the Examination of water and Wastewater, 19th Ed. American Public Health Association, Washington, D.C

3. American Public Health Association. *Standard Methods for the Examination of Water and Wastewater,* APHA, Washington, D.C.

4. Seyfried, Tobin, Brown and Ness. 1985. *Am. J. Public Health* ; 75:1071.

5. Kloos and Bannerman. 1999. *In* Murray, Baron, Pfaller, Tenover and Yolken (ed.), *Manual of Clinical Microbiology* , 7th ed. American Society for Microbiology, Washington, D.C.

Further Information

For further information please contact your local MICROMASTER Representative.

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

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

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