

MANNITOL SALT AGAR (MSA)

APPLICATION	Mannitol Salt Agar is used for selective isolation of pathogenic <i>Staphylococci</i> from pharmaceutical products in accordance with Microbial Limit Test by harmonized method of USP/EP														
PRINCIPLE AND INTERPRETATION	<p><i>Staphylococci</i> are widespread in nature, although they are mainly found on the skin, skin glands and mucous membranes of mammals and birds. The coagulase-positive species i.e. <i>Staphylococcus aureus</i> is well documented as a human opportunistic pathogen. <i>Staphylococci</i> have the unique ability of growing on a high salt containing media. Isolation of coagulase-positive <i>staphylococci</i> on Phenol Red Mannitol Agar supplemented with 7.5% NaCl was studied by Chapman . The resulting Mannitol Salt Agar Base is recommended for the isolation of coagulase positive <i>staphylococci</i> from cosmetics, milk, food and other specimens¹²³ The additional property of lipase activity of <i>Staphylococcus aureus</i> can be detected by the addition of the Egg Yolk Emulsion. The lipase activity can be visualized as yellow opaque zones around the colonies⁴. It is also used in the performance of microbial limit tests for the selective isolation of <i>Staphylococcus</i>. The formulation is in accordance with the harmonization of USP/EP⁵. The medium contains beef extract, pancreatic digest of casein and peptic digest of animal tissue which makes it very nutritious as they provide essential growth factors and trace nutrients. Many other bacteria except <i>Staphylococci</i> are inhibited by 7.5% sodium chloride. Mannitol is the fermentable carbohydrate fermentation of which leads to acid production, detected by phenol red indicator. <i>S.aureus</i> ferment mannitol and produce yellow coloured colonies surrounded by yellow zones. Coagulase-negative strains of <i>S.aureus</i> are usually mannitol non-fermenters and therefore produce pink to red colonies surrounded by red-purple zones. Presumptive coagulase-positive yellow colonies of <i>S.aureus</i> should be confirmed by performing the coagulase test [tube or slide] . Lipase activity of <i>S.aureus</i> can be detected by supplementing the medium with egg yolk emulsion. A possible <i>S.aureus</i> must be confirmed by the coagulase test. Also the organism should be subcultured to a less inhibitory medium not containing excess salt to avoid the possible interference of salt with coagulase testing or other diagnostic tests (e.g. Nutrient Broth⁶. Few strains of <i>S.aureus</i> may exhibit delayed mannitol fermentation. Negative results should therefore be re-incubated for an additional 24 hours before being discarded⁷.</p>														
MEDIUM COMPOSITION*	<table style="width: 100%; border-collapse: collapse;"> <tr> <td>Peptic Digest of animal tissue</td> <td style="text-align: right;">5.00 g/l</td> </tr> <tr> <td>Pancreatic digest of casein</td> <td style="text-align: right;">5.00g/l</td> </tr> <tr> <td>Beef Extract</td> <td style="text-align: right;">1.00 g/l</td> </tr> <tr> <td>Sodium chloride</td> <td style="text-align: right;">75.00 g/l</td> </tr> <tr> <td>D-Mannitol</td> <td style="text-align: right;">10.00 g/l</td> </tr> <tr> <td>Phenol Red</td> <td style="text-align: right;">0.025 g/l</td> </tr> <tr> <td>Agar</td> <td style="text-align: right;">15.000 g/l</td> </tr> </table> <p>Final pH 7.4± 0.2</p> <p>*Adjusted and/or supplemented to meet performances criteria</p>	Peptic Digest of animal tissue	5.00 g/l	Pancreatic digest of casein	5.00g/l	Beef Extract	1.00 g/l	Sodium chloride	75.00 g/l	D-Mannitol	10.00 g/l	Phenol Red	0.025 g/l	Agar	15.000 g/l
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STORAGE	+2°/+25°C Protect from light, excessive heat, moisture and freezing														

¹ Murray P.R., Baron J.H., Tenover F.C., Tenover F.C., Ed 2003, Manual of Clinical Microbiology, (thEd, American Society for Microbiology, Washington D.C.

² Koch P.K., 1942, Zentralbl., Bakteriolog., Parasitenkd. Abt I Orig. 149:122

³ Chapman G.H., 1945, J. Bacteriol., 50:201

⁴ Hitchins A.D., Tran T. and McCarron J.E., 1995, FDA Bacteriological Analytical Manual, 8th Ed. AOAC International, Gaithersburg, Md.

⁵ European Pharmacopoeia current Edition

⁶ Gunn B.A., Dunkelberg W.E. and Creitz J.R., 1972, Am. J. Clin. Pathol., 57:236

⁷ MacFaddin J.F., 1985, Media for Isolation-Cultivation-Identification-Maintenance of medical bacteria, vol. 1, Williams and Wilkins, Baltimore.

MANNITOL SALT AGAR (MSA)

QUALITY CONTROL	Growth Promotion Test: 10-100 viable microorganisms ⁸		
	Control strain	Incubation Conditions	Specifications
	<i>S. aureus</i> ATCC 6538	18 h at 32.5 ± 2.5°C	Good to luxuriant growth. Colonies yellow coloured
	<i>S. epidermidis</i> ATCC 12228	18 h at 32.5 ± 2.5°C	Good growth. Colonies red coloured
	<i>E. coli</i> ATCC 8739	72 h at 32.5 ± 2.5°C	Inhibited
	Sterility control		No growth
	Appearance		Red coloured, clear to slightly opalescent gel forms in plates
BARCODE	Data matrix code is composed of 20 digits: Digits 1→2 Media code Digits 3→7 Batch number Digits 8→9 Sub-batch number Digits 10→14 Progressive number Digits 15→20 Espiri Date (DDMMYY)		
GENERAL WARNING NOTES	Device must be handled according to asepsis precautions, of utilization of culture media is strictly referred to the type of analysis that must be done. Please refer to specific norms and procedures. Do not use if device is broken. Do not use if media shows accidental contamination signs. Do not utilize after expiry date. Let device reach room temperature before utilizing. Results interpretation must be done by qualified personnel, who must consider context of use. Disposal of waste must be carried out according to national and local regulations in force.		

This product is available in:

- **Non Gamma irradiated media plates**

MODEL	PRODUCT CODE	ORDER CODE	DESCRIPTION	SHELF LIFE
Ø90mm	MU118/10	MU118/10.100 (100 pcs/pack)	Filling volume: 24ml Packaging: single wrapped (S.W.)	4 months

⁸ For *E. coli* ≥100 viable microorganisms