

INSTRUCTIONS FOR USE – READY-TO-USE PLATED MEDIA

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PA-254027.07

BD™ Mannitol Salt Agar

INTENDED USE

BD Mannitol Salt Agar is used for the selective isolation of staphylococci and the detection of *Staphylococcus aureus* from clinical specimens.

PRINCIPLES AND EXPLANATION OF THE PROCEDURE

Microbiological method.

Mannitol Salt Agar is a formulation devised by Chapman for the differentiation of coagulase positive staphylococci (e.g., *Staphylococcus aureus*) from coagulase negative staphylococci. Mannitol Salt Agar is used for isolating staphylococci from clinical specimens, from cosmetics, and in microbial limit tests. 4,5

Mannitol Salt Agar contains peptones and beef extract, which supply essential nutrients. The 7.5% concentration of sodium chloride results in the partial or complete inhibition of bacterial organisms other than staphylococci. Mannitol fermentation, as indicated by a change in the phenol red indicator, aids in the differentiation of staphylococcal species. Coagulase positive staphylococci (e.g., *Staphylococcus aureus*) produce yellow colonies and a surrounding yellow medium while coagulase negative staphylococci produce red colonies and no colour change of the phenol red indicator.¹

REAGENTS

BD Mannitol Salt Agar

Formula* Per Liter Purified Water

Beef Extract	1.0 g
Pancreatic Digest of Casein	5.0
Peptic Digest of Animal Tissue	5.0
Sodium Chloride	75.0
D-Mannitol	10.0
Phenol Red	0.025
Agar	15.0

 $pH 7.4 \pm 0.2$

PRECAUTIONS

 \square . For professional use only. $^{\textcircled{2}}$

Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

Consult **GENERAL INSTRUCTIONS FOR USE** document for aseptic handling procedures, biohazards, and disposal of used product.

STORAGE AND SHELF LIFE

On receipt, store plates in the dark at 2 to 8° C, in their original sleeve wrapping until just prior to use. Avoid freezing and overheating. The plates may be inoculated up to the expiration date (see package label) and incubated for the recommended incubation times.

Plates from opened stacks of 10 plates can be used for one week when stored in a clean area at 2 to 8° C.

USER QUALITY CONTROL

Inoculate representative samples with the following strains (for details, see **GENERAL INSTRUCTIONS FOR USE** document). Incubate plates at 35 ± 2°C in an aerobic atmosphere.

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^{*}Adjusted and/or supplemented as required to meet performance criteria.

Examine plates after 18 to 24 and 48 h for amount of growth, colony size, pigmentation and selectivity. Typical reactions are as follows:

Strains	Growth results
Staphylococcus aureus ATCC™ 6538	Medium-sized yellow colonies, medium yellow
Staphylococcus aureus ATCC 25923	Medium-sized yellow colonies, medium yellow
Staphylococcus epidermidis ATCC 12228	Small to medium-sized white colonies, medium red
Proteus mirabilis ATCC 12453	Inhibition partial (to complete); colonies colorless, swarming inhibited
Escherichia coli ATCC 25922	Inhibition complete
Uninoculated	Red

PROCEDURE

Materials Provided

BD Mannitol Salt Agar (90 mm **Stacker**™ plates). Microbiologically controlled.

Materials Not Provided

Ancillary culture media, reagents and laboratory equipment as required.

Specimen Types

This is a selective differential medium for staphylococci that can be used for all clinical specimens (see also **PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE**). It is also used for non-clinical materials.

Test Procedure

Streak the specimen as soon as possible after it is received in the laboratory. The streak plate is used primarily to isolate pure cultures from specimens containing mixed flora. Alternatively, if material is being cultured directly from a swab, roll the swab over a small area of the surface at the edge and streak from this inoculated area. A nonselective medium such as Columbia Agar with 5% Sheep Blood must also be inoculated to provide an indication of other organisms present in the specimen.

Incubate plates at 24 to 48 h at $35 \pm 2^{\circ}$ C in an aerobic atmosphere.

Results

After incubation, plates are examined for the presence of staphylococcal colonies. Typical colonial morphology on **BD Mannitol Salt Agar** is as follows:

Organisms	Growth Results
Staphylococcus aureus	Medium-sized yellow colonies, medium yellow
Staphylococcus epidermidis	Small to medium white colonies, medium red
Staphylococci other than S. aureus	Small to large colonies with red or yellow zones,
and S. epidermidis	depending on the species
Micrococci	Large, white to orange
Enterococcus, Streptococcus	No growth to very weak growth
Gram-negative bacteria	No growth to weak growth

Colonies exhibiting the appearance of staphylococci must be further differentiated to confirm their identity.²

PERFORMANCE CHARACTERISTICS AND LIMITATIONS OF THE PROCEDURE

BD Mannitol Salt Agar is a standard formulation used for the isolation and differentiation by mannitol fermentation of staphylococci from clinical and non-clinical sources. Incubation periods of 48 to 72 hours are recommended to detect all staphylococcal species present in the specimen.²

Several *Staphylococcus* species other than *S. aureus* are mannitol positive and produce yellow colonies surrounded by yellow zones on this medium (e.g. *S. capitis, S. xylosus, S. cohnii, S. sciuri, S. simulans*, and other species). Therefore, further biochemical tests are necessary for the identification of *S. aureus* or other species. Consult appropriate references. ^{2,3}

REFERENCES

- 1. Chapman, G.H. 1945. The significance of sodium chloride in studies of staphylococci. J. Bacteriol. 50:201-203.
- Bannerman, T.L. 2003. Staphylococcus, Micrococcus, and other catalase-positive cocci that grow aerobically. In: Murray, P. R., E. J. Baron, J.H. Jorgensen, M. A. Pfaller, and R. H. Yolken (ed.). Manual of clinical microbiology, 8th ed. American Society for Microbiology, Washington, D.C.
- 3. Hitchins, A. D., T. T. Tran, and J. E. McCarron. 1995. Microbiology methods for cosmetics, p. 23.01-23.12. In Bacteriological analytical manual, 8th ed. AOAC International, Gaithersburg, MD.
- 4. U.S. Pharmacopeial Convention, Inc. The U.S. Pharmacopeia /The national formulary. *Current edition*. U.S. Pharmacopeial Convention, Inc., Rockville, Md. USA
- 5. Council of Europe. European Pharmacopoeia, *current edition*. European Pharmacopoeia Secretariat. Strasbourg/France.

PACKAGING/AVAILABILITY

BD Mannitol Salt Agar

Cat. No. 254027 Ready-to-use plated media, 20 plates Cat. No. 254079 Ready-to-use plated media, 120 plates

FURTHER INFORMATION

For further information please contact your local BD representative.



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