

MANNITOL SALT AGAR (7143)

Intended Use

Mannitol Salt Agar is used for the isolation of staphylococci. Conforms to Harmonized USP/EP/JP Requirements.^{1,2,3}

Product Summary and Explanation

Chapman formulated Mannitol Salt Agar to isolate staphylococci by inhibiting growth of most other bacteria with a high salt concentration.⁴ Chapman added 7.5% Sodium Chloride to Phenol Red Mannitol Agar, and noted pathogenic strains of staphylococci (coagulase-positive staphylococci) grew luxuriantly and produced yellow colonies with yellow zones. Nonpathogenic staphylococci produced small red colonies with no color change to the surrounding medium.

Mannitol Salt Agar is highly selective, and specimens from heavily contaminated sources may be streaked onto this medium without danger of overgrowth.⁵ Mannitol Salt Agar is recommended for isolating pathogenic staphylococci from clinical specimens, cosmetics, and microbial limit tests.^{1,2,3,5,6}

Principles of the Procedure

Enzymatic Digest of Casein, Enzymatic Digest of Animal Tissue, and Beef Extract provide the nitrogen, vitamins, and carbon in Mannitol Salt Agar. D-Mannitol is the carbohydrate source. In high concentrations, Sodium Chloride inhibits most bacteria other than staphylococci. Phenol Red is the pH indicator. Agar is the solidifying agent.

Bacteria that grow in the presence of a high salt concentration and ferment mannitol produce acid products, turning the Phenol Red pH indicator from red to yellow. Typical pathogenic staphylococci ferment mannitol and form yellow colonies with yellow zones. Typical non-pathogenic staphylococci do not ferment mannitol and form red colonies.

Formula / Liter

Enzymatic Digest of Casein	5 g
Enzymatic Digest of Animal Tissue	5 g
Beef Extract.....	1 g
D-Mannitol	10 g
Sodium Chloride.....	75 g
Phenol Red.....	0.025 g
Agar.....	15 g

Final pH: 7.4 ± 0.2 at 25°C

Formula may be adjusted and/or supplemented as required to meet performance specifications.

Precautions

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

Directions

1. Suspend 111 g of the medium in one liter of purified water.
2. Heat with frequent agitation and boil for one minute to completely dissolve the medium.
3. Autoclave at 121°C for 15 minutes.

Quality Control Specifications

Dehydrated Appearance: Powder is homogeneous, free flowing, and light red-orange to light beige.

Prepared Appearance: Prepared medium is trace to slightly hazy and peach to pink.

Expected Cultural Response and USP/EP/JP Growth Promotion Testing: Cultural response on Mannitol Salt Agar incubated at Harmonized USP/EP/JP specified temperatures and incubation times.^{1,2,3}

Microorganism	Approx. Inoculum (CFU)	Expected Results	
		Recovery	Reactions
<i>Staphylococcus aureus</i> ATCC® 6538	10 - 100	Fair to good	Yellow colonies; may have yellow halo around colonies.
<i>Staphylococcus aureus</i> ATCC® 25923	10 - 100	Fair to good	Yellow colonies; may have yellow halo around colonies
<i>Staphylococcus epidermidis</i> ATCC® 12228	10 - 100	Fair to good	Colorless to pink colonies
<i>Escherichia coli</i> ATCC® 8739	300 - 1000	Inhibited	---

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

Test Procedure

Inoculate specimen on medium as a primary isolation or inoculate isolated colonies onto medium for differentiation.

Results

Staphylococci will grow on this medium, while the growth of most other bacteria will be inhibited. Coagulase-positive staphylococci will produce luxuriant growth of yellow colonies and may have a yellow halo around the colony. Coagulase-negative staphylococci will produce small colorless to pink colonies with no color change to the medium.

Storage

Store 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 by keeping container tightly closed.

Expiration

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

Limitation of the Procedure

Due to nutritional variation, some strains may be encountered that grow poorly or fail to grow on this medium.

Packaging

Mannitol Salt Agar	Code No.	7143A	500 g
		7143B	2 kg
		7143C	10 kg

References

1. **United States Pharmacopeial Convention.** 2007. The United States pharmacopeia, 31st ed., Amended Chapters 61, 62, 111. The United States Pharmacopeial Convention, Rockville, MD.
2. **Directorate for the Quality of Medicines of the Council of Europe (EDQM).** 2007. The European Pharmacopoeia, Amended Chapters 2.6.12, 2.6.13, 5.1.4, Council of Europe, 67075 Strasbourg Cedex, France.
3. **Japanese Pharmacopoeia.** 2007. Society of Japanese Pharmacopoeia. Amended Chapters 35.1, 35.2, 7. The Minister of Health, Labor, and Welfare.
4. **Chapman, G. H.** The significance of sodium chloride in studies of staphylococci. *J. bacteriol.* **50**:201.
5. **Kloos, W. E., and T. L. Bannerman.** 1995. *Staphylococcus* and *Micrococcus*. In P. R. Murray, E. J. Baron, M. A. Pfaller, F. C. Tenover, and R. H. Tenover (eds.). *Manual of clinical microbiology*, 6th ed. American Society for Microbiology, Washington, D.C.
6. **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.

Technical Information

Contact Acumedia Manufacturers, Inc. for Technical Service or questions involving dehydrated culture media preparation or performance at (517)372-9200 or fax us at (517)372-2006.