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Instructions for Use

MANNITOL SALT AGAR (MSA) WITH OXACILLIN

Cat. no. G97	Mannitol Salt Agar (MSA) with Oxacillin, 15x100mm Plate, 18ml	10 plates/bag

INTENDED USE

Hardy Diagnostics Mannitol Salt Agar (MSA) with Oxacillin is used as a primary screening medium for the simultaneous detection and differentiation of methicillin resistant *Staphylococcus* spp. (MRS). It can be used as a cost effective screening method for MRSA.

SUMMARY

Mannitol Salt Agar (MSA) is used as a selective, differential media for pathogenic staphylococci. Oxacillin has been added for selective isolation of methicillin-resistant strains. Oxacillin is used instead of methicillin due to its greater stability. This plate may be used to screen environmental and clinical specimens.

FORMULA

Ingredients per liter of deionized water:*

Sodium Chloride	60.0gm
Phenol Red Broth Base	16.0gm
Mannitol	10.0gm
Polymyxin B	10.0mg
Oxacillin	4.0mg
Agar	15.0gm

Final pH 7.4 +/- 0.2 at 25ºC.

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

STORAGE AND SHELF LIFE

Storage: Upon receipt store at 2-8ºC. away from direct light. Media should not be used if there are any signs of deterioration (shrinking, cracking, or discoloration), contamination, or if the expiration date has passed. Product is light and temperature sensitive; protect from light, excessive heat, moisture, and freezing.

The expiration date on the product label applies to the product in its intact packaging when stored as directed. The product may be used and tested up to the expiration date on the product label and incubated for the recommended

incubation times as stated below.

Refer to the document "Storage" for more information.

PRECAUTIONS

This product may contain components of animal origin. Certified knowledge of the origin and/or sanitary state of the animals does not guarantee the absence of transmissible pathogenic agents. Therefore, it is recommended that these products be treated as potentially infectious, and handle observing the usual Universal Precautions for blood. Do not ingest, inhale, or allow to come into contact with skin.

This product is for *in vitro* diagnostic use only. It is to be used only by adequately trained and qualified laboratory personnel. Observe approved biohazard precautions and aseptic techniques. All laboratory specimens should be considered infectious and handled according to "standard precautions." Refer to the document "Guidelines for Isolation Precautions" from the Centers for Disease Control and Prevention.

For additional information regarding specific precautions for the prevention of the transmission of all infectious agents from laboratory instruments and materials, and for recommendations for the management of exposure to infectious disease, refer to CLSI document M29: *Protection of Laboratory Workers from Occupationally Acquired Infections.*

Sterilize all biohazard waste before disposal.

Refer to the document "Precautions When Using Media" for more information.

PROCEDURE

Specimen Collection: Infectious material should be submitted directly to the laboratory without delay and protected from excessive heat and cold. If there is to be a delay in processing, specimens should be inoculated into the appropriate transport media and refrigerated until inoculation in a sterile container, or other appropriate means of transport. Consult appropriate references for specimen collection and transport.[1]

Method of Use: Prior to inoculation, the medium should be brought to room temperature. Inoculate the media with the specimen and streak for isolation. Incubate aerobically at 35ºC. for 24-48 hours.

INTERPRETATION OF RESULTS

Most methicillin-resistant *Staphylococcus aureus* (MRSA) are capable of fermenting mannitol within 24 hours. Fermentation of mannitol is indicated by a color change from red to yellow. However, delayed fermentation of mannitol may occur with a few strains of MRSA, so negative plates should be incubated for an additional 24 hours. It is recommended that a coagulase or latex agglutination test be performed on mannitol-fermenting isolates for a presumptive identification of MRSA.

MRS organisms other than *Staphylococcus aureus* may grow on the media in 24-48 hours but appear as mannitol non-fermentors and are coagulase-negative.

LIMITATIONS

It is recommended that biochemical, immunological, molecular, or mass spectrometry testing be performed on colonies from pure culture for complete identification of bacteria and/or fungi.

While the medium is differential and selective for MRSA, a coagulase or latex agglutination test as well as other antimicrobial and biochemical tests must be performed for complete identification.

Methicillin-resistant, coagulase-negative staphylococci will also grow on this medium but can be differentiated by their inability to ferment mannitol and a negative coagulase reaction.

Refer to the document "Limitations of Procedures and Warranty" for more information.

MATERIALS REQUIRED BUT NOT PROVIDED

Standard microbiological supplies and equipment such as loops, other culture media, swabs, applicator sticks, incinerators, and incubators, etc., as well as serological and biochemical reagents, are not provided.

QUALITY CONTROL

Hardy Diagnostics tests each lot of commercially manufactured media using appropriate quality control microorganisms and quality specifications as outlined on the Certificate of Analysis (CofA) and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media*. The following microorganisms are routinely used for testing at Hardy Diagnostics:

Test Organisms	Inoculation Method*	Incubation			Results
		Time	Temperature	Atmosphere	
Staphylococcus aureus ATCC® 43300	A	24-48hr	35°C	Aerobic	Growth; small colonies with yellow color change in media
Proteus mirabilis ATCC® 12453	A	24hr	35°C	Aerobic	Partial to complete inhibition
Staphylococcus aureus ATCC® 29213	B	24hr	35°C	Aerobic	Inhibited

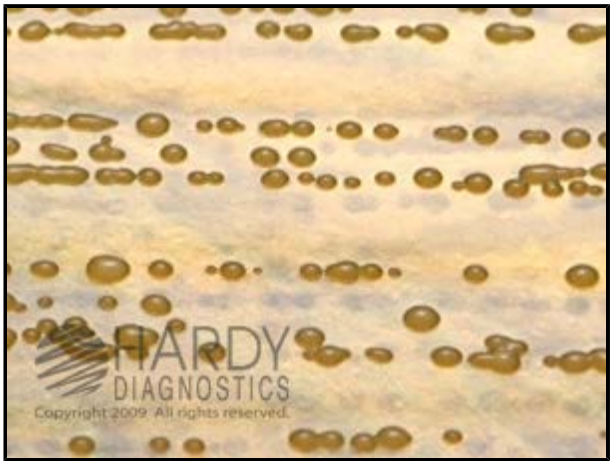
* Refer to the document "Inoculation Procedures for Media QC" for more information.

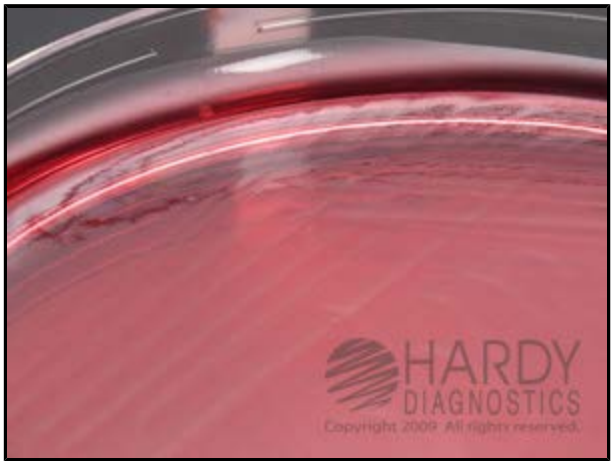
USER QUALITY CONTROL

End users of commercially prepared culture media should perform QC testing in accordance with applicable government regulatory agencies, and in compliance with accreditation requirements. Hardy Diagnostics recommends end users check for signs of contamination and deterioration and, if dictated by laboratory quality control procedures or regulation, perform quality control testing to demonstrate growth or a positive reaction and to demonstrate inhibition or a negative reaction, if applicable. Hardy Diagnostics quality control testing is documented on the certificate of analysis (CofA) available from Hardy Diagnostics Certificate of Analysis website. Also refer to the document "Finished Product Quality Control Procedures," and the CLSI document M22-A3 *Quality Assurance for Commercially Prepared Microbiological Culture Media* for more information on the appropriate QC procedures. See the references below.

PHYSICAL APPEARANCE

Mannitol Salt Agar (MSA) with Oxacillin should appear clear, and light orange in color.





Staphylococcus aureus (ATCC® 43300) colonies growing on Mannitol Salt Agar with Oxacillin (Cat. no. G97). Incubated aerobically for 48 hours at 35ºC.

Proteus mirabilis (ATCC® 12453) growth inhibited on Mannitol Salt Agar with Oxacillin (Cat. no. G97). Incubated aerobically for 48 hours at 35ºC.

REFERENCES

1. Jorgensen., et al. *Manual of Clinical Microbiology,* American Society for Microbiology, Washington, D.C.

2. Tille, P., et al. *Bailey and Scott's Diagnostic Microbiology,* C.V. Mosby Company, St. Louis, MO.

3. Lally, R.T., T.M. Ederer and B. Woolfrey. 1985. Evaluation of Mannitol Salt Agar and Oxacillin as a screening medium for methicillin-resistant *Staphylococcus aureus*. *J. Clin. Microbiol.*; 22: 501-504.

4. *Quality Assurance for Commercially Prepared Microbiological Culture Media*, M22. Clinical and Laboratory Standards Institute (CLSI - formerly NCCLS), Wayne, PA.

5. Van Enk, R.A. and K. Thompson. 1992. Primary isolation medium for the recovery of methicillin-resistant *Staphylococcus aureus*. *J. Clin. Microbiol.*; 30: 504-505.

6. Isenberg, H.D. *Clinical Microbiology Procedures Handbook,* Vol. I, II & III. American Society for Microbiology, Washington, D.C.

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The Hardy Diagnostics manufacturing facility and quality management system is certified to ISO 13485.

HDQA 2207F [D]