Reference: 100560ZA **Technical Data Sheet**

Product: **Mannitol Salt Agar**

Specification

Selective medium for the isolation of pathogenic staphylococci, according to the Pharmacopoeial Harmonized Methodology and Clinical samples.

Presentation

	Packaging Details	Shelf Life	Storage
20 Prepared Plates 90 mm with: 21 ± 2 ml	1 box with 2 packs of 10 plates/pack. Single cellophane.	3 months	2-14 ºC

Composition

Composition (g/l):

Meat extract	1.000
Casein peptone	5.000
Meat peptone	5.000
Sodium chloride	75.000
D-Mannitol	10.000
Phenol red	0.025
Agar	15.000

Description /Technique

Description:
Mannitol Salt Agar is a classical medium for the detection and enumeration of staphylococci. It was described by Chapman and has been adopted by many official organisations. Several modifications of it have been developed, all formulations resulting in media with similar efficiency.
This medium takes advantage of the high saline tolerance of staphylococci, and uses sodium chloride as a selective agent. Only staphylococci and halophilic enterobacteria are able to grow freely at the concentration of salt employed in this medium, while other bacteria are inhibited. It also exploits the correlation between the pathogenicity of staphylococci and their ability ferment mannitol. Mannitol fermentation results in an accumulation of acid products, indicated by the phenol red indicator turning yellow. A yellow halo surrounds the presumptive pathogenic colonies, while the rest of the medium remains red/orange in colour.

Technique:
Inoculate the plates and incubate at 37 °C for 36 hours or at 30-35 °C for 3 days.

The typical appearance of the colonies after the correct incubation is as follows:
- Presumptive pathogenic staphylococci (coagulase +) are mannitol positive and produce large colonies with a yellow halo.
- Non-pathogenic staphylococci (coagulase -) are usually mannitol negative and produce small colonies without a halo or change in colour.

Coagulase presence must be tested by the classical technique in order to establish its true pathogenic potential.

Note: According to the methodology chosen by the laboratory (Pharmacopeia or other international standards), may be slight variations in incubation times and temperatures, as well as inhibition of *E. coli*, which can be variable depending on the inoculated bacterial population . This medium can normally reduce the bacterial load by up to 3 decimal logarithms.

Precautions
For in vitro diagnostic use. Do not reuse. For professional use only.
Do not use the product if it shows evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

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Quality control

Physical/Chemical control

Color : Strongly pink pH: 7.4 ± 0.2 at 25ºC

Microbiological control

Inoculate with 10-100 CFU according to harmonized Pharmacopoeia or with 100-1000 CFU for selectivity.

Analytical methodology according to ISO 11133:2014/A1:2018; A2:2020.

Microbiological control according to ISO 11133:2014/A1:2018; A2:2020.

Aerobiosis. Incubation at 30-35ºC. Reading at 24h (qualitative productivity), 48h (quantitative productivity) and 72h (Selectivity)

Microorganism	Growth
Escherichia coli ATCC® 8739, WDCM 00012	Inhibited
Stph. epidermidis ATCC® 12228, WDCM 00036	Poor to good- White colonies -Red medium
Staphylococcus aureus ATCC® 6538, WDCM 00032	Good (≥ 50%). Yellow colonies. Yellow medium.
Stph. aureus ATCC® 25923, WDCM 00034 (24h)	Good
Stph. aureus ATCC® 25923, WDCM 00034 (48h)	Good (≥ 50%). White colonies. Yellow medium.

Sterility Control

Incubation 48 h at 30-35 ºC and 48 h at 20-25 ºC: NO GROWTH.
Check at 7 days after incubation in same conditions.

Bibliography

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· DOWNES, F.P. & K. ITO (2001) Compendium of Methods for the Microbiological Examination of Foods. 4th ed. APHA. Washington. DC. USA.
· EUROPEAN PHARMACOPOEIA 11.0 (2023) 11th ed. § 2.6.13. Microbiological examination of non-sterile products: Test for specified microorganisms. Harmonised Method. EDQM. Council of Europe. Strasbourg.
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. ISO 11133:2014/ Adm 1:2018. Microbiology of food, animal feed and water. Preparation, production, storage and performance testing of culture media.
· ISO 22718 Standard (2015) . Cosmetics - Microbiology - Detection of Staphylococcus aureus.
· USP 33 - NF 28 (2011) <62> Microbiological examination of non-sterile products: Test for specified microorganisms. Harmonised Method. USP Corp. Inc. Rockville. MD. USA.

Storage

Storage conditions: 2-14ºC
Alternatively the plates may also be stored at the range of 2 - 25ºC, with a proper performance of the medium, but some precautions must be taken into account:
-In the range of 2 - 8 ºC avoid direct contact with surfaces that can freeze product.
-In the range of 15 - 25 ºC, dehydration control must be taking in account.

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