M-Staphylococcus Broth

M1120

Intended Use:

Recommended for detection and isolation of Staphylococci by membrane filtration technique.

Composition**

Ingredients	g / L
Tryptone	10.000
Yeast extract	2.500
Lactose	2.000
Mannitol	10.000
Dipotassium hydrogen phosphate	5.000
Sodium chloride	75.000
Sodium azide	0.049
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 104.55 grams in 1000ml purified/distilled water.Mix thoroughly and boil for 5 minutes. DO NOT AUTOCLAVE. For 10 ml inocula use double strength medium.

Principle And Interpretation

The swimming pool water is generally potable and treated with additional disinfectants but it also may come from thermal springs or salt water. Modern pools have a recirculation system for filtration and disinfection. Staphylococci are gram-positive cocci residing on the skin and mucous membrane of humans and other organisms. M-Staphylococcus Broth is used for detection and isolation of Staphylococci by membrane filter technique. This broth is especially used for isolating pathogenic and enterotoxigenic Staphylococci and has similar composition as Staphylococcus Agar No. 110 except agar and gelatin (1).

Tryptone and yeast extract supply essential growth factors such as nitrogen, carbon, sulphur, vitamins and trace ingredients. The 7.5% concentration of sodium chloride results in partial or complete inhibition of bacteria except Staphylococci. Mannitol and lactose are utilized as energy sources.

Type of specimen

Water samples

Specimen Collection and Handling:

Inoculate the tubes of M-Staphylococcal Broth and incubate at 35 ± 2°C for 24 hours. Streak from positive tubes (turbid growth) on plates of Lipovitellin Salt Mannitol Agar Base (M627) and incubate at 35-37°C for 48 hours. Opaque, yellow zones around the colonies are positive evidence of lipovitellin- lipase activity and mannitol fermentation (2). Alternatively around 2 ml of M-Staphylococcus Broth is used to saturate sterile absorbent cotton pads. Membrane filters used for filtration are aseptically placed on these saturated cotton pads. Following an incubation at 35-37°C for 18-48 hours, observe membrane for growth and pigment production. Mannitol fermentation can be visualized as yellow colouration by addition of a few drops of bromothymol blue to the areas from where colonies have been removed. After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations

1. Further biochemical and serological tests must be carried out for further identification.
2. Individual organisms differ in their growth requirement and may show variable growth patterns on the medium.
3. Each lot of the medium has been tested for the organisms specified on the COA. It is recommended to users to validate the medium for any specific microorganism other than mentioned in the COA based on the user's unique requirement.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity

Light amber coloured clear solution without any precipitate

Reaction

Reaction of 10.45% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH

6.80-7.20

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth
Enterococcus faecalis ATCC 29212 (00087*)	>=10^4	inhibited
Escherichia coli ATCC 25922 (00013*)	>=10^4	inhibited
Staphylococcus aureus subsp. *aureus* ATCC 25923 (00034*)	50-100	good-luxuriant
Staphylococcus epidermidis ATCC 12228 (00036*)	50-100	good-luxuriant
Streptococcus pyogenes ATCC 19615	>=10^4	inhibited

Key : *Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition. Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Maintenance-of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
2. Lipps WC, Braun-Howland EB, Baxter TE,eds. Standard methods for the Examination of Water and Wastewater, 24th ed. Washington DC:APHA Press; 2023.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.

Revision : 03/2024