



M-Staphylococcus Broth

M1120

M-Staphylococcus Broth is used for detection and isolation of Staphylococci by membrane filter technique.

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	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 1000 ml distilled water. Mix thoroughly and heat to boiling for 5 minutes. DO NOT AUTOCLAVE. For 10 ml inocula, use double strength medium.

Warning : Sodium azide has a tendency to form explosive metal azides with plumbing materials. It is advisable to use enough water to flush off the disposables.

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).

Casein enzymic hydrolysate 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.

Inoculate the tubes of M-Staphylococcal Broth and incubate at $35 \pm 2^\circ\text{C}$ for 24 hours. Streak from positive tubes (turbid growth) on plates of Lipovitellin Salt Mannitol Agar Base (M627) and incubate at $35-37^\circ\text{C}$ 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 $35-37^\circ\text{C}$ 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.

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

M1120: Cultural characteristics observed after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth
Cultural Response		
<i>Enterococcus faecalis</i> ATCC 29212	$\geq 10^3$	inhibited
<i>Escherichia coli</i> ATCC 25922	$\geq 10^3$	inhibited
<i>Staphylococcus aureus</i> ATCC 25923	50-100	good-luxuriant
<i>Staphylococcus epidermidis</i> ATCC 12228	50-100	good-luxuriant
<i>Streptococcus pyogenes</i> ATCC 19615	$\geq 10^3$	inhibited

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference

1. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Maintenance-of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.
2. Eaton A. D., Clesceri L. S. and Greenberg A. E., (Eds.), 1995, Standard Methods for the Examination of water and Wastewater, 19th Ed. American Public Health Association, Washington, D.C.

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