Staphylococcus Agar No 110 with Azide

M156

Staphylococcus Agar No.110 is used as a selective medium for the isolation and testing of pathogenic *Staphylococci* .

Composition**

Ingredients	Gms / Litre
Casein enzymic hydrolysate	10.000
Yeast extract	2.500
Gelatin	30.000
Lactose	2.000
D-Mannitol	10.000
Sodium chloride	75.000
Dipotassium phosphate	5.000
Sodium azide	0.100
Agar	15.000
Final pH (at 25°C)	7.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 149.6 grams in 1000 ml of warm distilled water. Mix thoroughly. Heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Resuspend the precipitate by gentle agitation to avoid bubbles and pour the plates while the medium is hot. Alternatively, cool the medium to 45 - 50°C and add blood or egg yolk if desired.

Caution : 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

Staphylococcus Agar No. 110 is formulated as described by Chapman (1, 2, 3) for selective isolation and enumeration of *Staphylococci* from clinical as well as nonclinical specimens. Staphylococcus Agar No. 110 with azide is used for determination of coagulase positive *Staphylococci* in meat pies even in the presence of large number of *Bacillus* species (4). This medium is recommended by APHA (5). The addition of blood in the medium enables to study haemolytic reaction (6) and with egg yolk enables to study lecithinase production by *Staphylococcus aureus* (7). This medium is selective due to high salt concentration and differential on the basis of ability of organism to ferment mannitol, produce pigment and gelatin liquefaction.

This medium is very nutritive as it contains Casein enzymic hydrolysate and yeast extract which provide essential growth factors like vitamins, nitrogen, carbon compounds, sulphur and trace nutrients etc. to the organisms. High concentration of sodium chloride inhibits many bacterial species except *Staphylococci.* Sodium azide inhibits gram-negative organisms. Mannitol fermentation can be visualized as yellow colouration by addition of a few drops of bromo thymol blue to the areas of the plates from where colonies have been removed. Gelatin liquefaction can be seen when the plates are flooded with a saturated aqueous solution of ammonium sulphate. *Enterococcus faecalis* may grow on this medium as small colonies with little mannitol fermentation (8).

Quality Control

Appearance
Cream to yellow homogeneous free flowing powder

Gelling
Firm, comparable with 1.5% Agar gel and 3.0% gelatin gel

Colour and Clarity of prepared medium
Light amber coloured clear to slightly opalescent gel forms in Petri plates

Reaction
Reaction of 14.96% w/v aqueous solution at 25°C. pH : 7.0±0.2

pH
6.80-7.20

Cultural Response
M156: Cultural characteristics observed after an incubation at 35-37°C for 48 hours .

Organism	Inoculum (CFU)	Growth	Recovery	Mannitol fermentation (on addition of BTB)	Pigment Production	Gelatinase production (flooding plate with standard aqueous solution of ammonium sulphate)
Staphylococcus aureus ATCC 25923	50-100	good-luxuriant	>=50%	positive reaction	positive	positive reaction
Staphylococcus epidermidis ATCC 12228	50-100	good-luxuriant	>=50%	variable reaction	negative	positive reaction
Enterococcus faecalis ATCC 29212	50-100	none-poor	<=10%	slight reaction	negative	variable reaction
Escherichia coli ATCC 25922	>=10^3	inhibited	0%			

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. Chapman G.H., 1946, J. Bact., 51:409.
2. Chapman G.H., 1948, Food Res., 13:100.
3. Chapman G.H., 1952, J. Bact., 63:147.
4. Smucker S.A. and Appleman M.D., 1964, Appl. Microbiol., 12(4):355.
5. Speck M. (Ed.), 1984, Compendium of Methods for the Microbiological Examination of Foods, 2nd ed., APHA, Washington, D.C.
6. Shaffer J. C. and McDade J. J., 1962, Arch. Environ. Health, 5:547.
7. Carter C.H., 1960, J. Bact., 79:753.
8. MacFaddin J., 1985, Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.

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