70193 Staphylococcus Agar (CHAPMAN-Agar; Staphylococcus Medium No. 110)
NutriSelect® Plus

Selective medium for isolation and differentiation of pathogenic staphylococci on the basis of salt tolerance, pigmentation, D-mannitol utilization and gelatin liquefaction. Smuckler and Appleman recommend addition of sodium azide in order to make this medium selective for the determination of coagulase-positive staphylococci.

Composition:

Ingredients	Grams/Litre
Yeast extract	2.5
Tryptone	10.0
Lactose	2.0
Mannitol	10.0
Sodium chloride	75.0
Dipotassiumhydrogen phosphate	5.0
Gelation	30.0
Agar	15.0

Final pH 7.1 +/- 0.2 at 37°C

Store dehydrated powder between 10-30°C in a tightly closed container and the prepared medium at 2-8°C. Protect from moisture and light by keeping container in a low humidity environment.

Appearance(color):	Faintly yellow & Faint beige & Faint brown, free flowing powder
Gelling:	Firm, comparable with 1.5% Agar gel and 3.0% gelatin gel
Color and Clarity:	Light amber coloured clear to slightly opalescent gel forms in Petri plates

Directions:
Dissolve 149.5 g in 1 litre distilled water and sterilize by autoclaving at 121°C for 15 minutes.

Principle and Interpretation:
Staphylococcus Agar No. 110 (1,3,4) also known as Stone Gelatin Agar (5) is used for the selective isolation of pathogenic Staphylococci on the basis of salt tolerance, pigment production, mannitol fermentation and gelatin liquefaction. Pathogenic staphylococci (coagulase-positive) are able to grow on the high-salt mannitol medium to form orange colonies which give positive reactions for acid production and gelatin liquefaction. These properties are few of the characteristics of pathogenic Staphylococci (7, 8).
Smuckler & Appleman(6)made Staphylococcus Medium No.110 selective, for the determination of coagulase-positive staphylococci in meat pies containing large numbers of *Bacillus* spp., by the addition of sodium azide 0.75 mM (4.875g/L). Staphylococcus Agar No. 110 is recommended by APHA (1) and AOAC (2). The medium can be used with Egg Yolk Emulsion (17148) to study the egg yolk reactions (9).
Tryptone and yeast extract serve as sources of carbon, nitrogen and other essential nutrients and growth factors including vitamins. D-Mannitol is the fermentable carbohydrate with lactose being an additional source of carbon. Sodium chloride maintains the osmotic equilibrium while phosphate buffers the medium. Gelatin serves as the substrate for gelatin liquefaction.
Mannitol fermentation can be visualized as yellow colouration by addition of a few drops of bromothymol blue to the areas of the plates where colonies have been removed. Gelatin hydrolysis may be demonstrated by adding a drop of a saturated aqueous solution of ammonium sulphate or,

preferably, of a 20% aqueous solution of sulphosalicylic acid to an individual colony ('Stone reaction'). A positive `Stone reaction' is denoted by the presence of a clear zone round gelatinase-producing colonies after 10 minutes' contact with the reagent.

Cultural characteristics after 48 hours at 35-37°C. (Mannitol fermentation - on addition of BTB; Gelatinase production: flooding plate with standard aqueous solution of ammonium sulphate)

Organisms (ATCC/WDCM)	Inoculum (CFU)	Growth	Recovery	Mannitol Fermentation	Pigment production	Gelatinase production
Staphylococcus aureus subsp. aureus (25923/ 00034)	50-100	++/+++	≥50%	+	+	+
Staphylococcus epidermidis (12228/00036)	50-100	++/+++	≥50%	variable reaction	-	+
Enterococcus faecalis (29212/00087)	50-100	+/-	≤10%	Slight reaction	-	variable reaction
Escherichia coli (25922/00013)	$\geq 10^4$	-	0%			

References:

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2. Association of Official Analytical Chemists (AOAC), Bacteriological Analytical Manual, 5th Ed., 1978, AOAC International, Gaithersburg, Md.
3. Baird R.B., Eaton A.D., and Rice E.W., (Eds.), 2015, Standard Methods for the Examination of Water and Wastewater, 23rd ed., APHA, Washington, D.C.
4. Carter C. H., 1960, J. Bacteriol., 79:753.
5. Stone R. V., 1935. Proc. Soc. Exper. Biol. and Med. 33:185-187.
6. BD Smuckler S. A. and Appleman M. D. (1964) *Appl. Microbiol*. 12. 335-339
7. Chapman G. H., 1947, J. Bacteriol., 53:504.
8. Chapman G. H., 1952, J. Bacteriol., 63:147
9. 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.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.