CHAPMAN STONE AGAR TM 061

For selective isolation of Staphylococci associated with food poisoning

Composition

Ingredients	Gms/Ltr.
Ammonium sulphate	75.00
Sodium chloride	55.00
Gelatin	30.00
Agar	15.00
Casein peptone	10.00
Mannitol	10.00
Yeast extract	2.50
Dipotassium phosphate	5.00

* Dehydrated powder store, hygroscopic in nature, place in a dry, in tightly - sealed containers below 25°C and protect from direct Sunlight.

Instructions for Use

Dissolve 202.5gms in 1000ml distilled water. Gently heat to boiling with gentle swirling and dissolve the medium completely. Sterilize at 15 psi (121°C) for 10 minutes. Cool at 45 - 50°C and dispense into sterile petri plates.

Appearance: Light to medium amber, opalescent with ppt. gel
pH (at 25°C): 7.0 ± 0.2

Principle

CHAPMAN STONE AGAR is used for selective isolation of Staphylococci associated with food poisoning(1). Medium is recommended by SMUCKLER and APPLEMAN (1964). Medium contains Casein peptone provides nitrogen, vitamins, minerals and amino acids essential for growth. Yeast extract is a source of vitamins, particularly of the B-group essential for bacterial growth. Mannitol is a fermentable carbohydrate source providing carbon and energy and its fermentation can be detected by adding a few drops of bromocresol purple resulting in production of yellow colour. Addition of high concentrations of Sodium chloride helps in inhibiting most bacteria except Staphylococci; Gelatin is a protein derived by the hydrolysis of collagen, found abundantly in bones, skin, tendons, cartilage and animal tissue. It is used in culture media to determine gelatinolysis by bacteria. Gelatin hydrolysis is observed as clear zones around colonies. Due to the presence of ammonium sulphate in the medium itself there is no need to flood the plate with ammonium sulphate solution for detection of gelatin liquefaction by the isolates, which is known as Stones method (2). Dipotassium phosphate provides buffering capability. Material under test is inoculated on the surface and incubated at 30°C for 48 hours to produce separated colonies. After incubation, cream to golden yellow colonies surrounded by clear zones are presumptively identified as S. aureus . White or non-pigmented colonies, with or without a clear zone, are presumptively identified as S. epidermidis . Coagulase activity should be performed to confirm the findings.
Enterococci and/or Group D streptococci may exhibit growth on the medium and show slight mannitol fermentation. The colonies, however, are tiny and can easily be differentiated from staphylococci by gram stain and the catalase test (3)

Interpretation

Cultural characteristics observed after inoculating (10^2 - 10^3CFU/ml), on incubation at 30 ± 2°C for 18 - 48 hours at aerobically.

Microorganisms	ATCC	Inoculum (CFU/ml)	Growth	Mannitol fermentation	Halo
Escherichia coli	25922	10^2 - 10^3	Inhibited	NA	NA
Staphylococcus epidermidis	12228	10^2 - 10^3	Luxuriant	-ve reaction, no production of yellow colour on addition of Bromo cresol purple	+ve reaction
Satphylococcus aureus	25923	10^2 - 10^3	Luxuriant	+ve reaction, production of yellow colour on addition of Bromo cresol purple	+ve reaction

References

1. Chapman J. Bact. 1945. 50: 201 Recommended Methods for the Microbiological Examination of Foods APHA. Inc. New York. (1958).
2. Stone, 1935, Proc. Soc. Exp. Biol. N.Y., 33:185.
3. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Identification -Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore