TMP 017 – SHEEP BLOOD AGAR PLATE

INTENDED USE

For cultivation of fastidious organisms and studying haemolytic reactions.

PRODUCT SUMMARY AND EXPLANATION

Sheep Blood Agar medium is both differential and enriched medium and used for the isolation, cultivation and detection of hemolytic activity of fastidious microorganisms like Streptococci and Pneumococci. The Blood Agar plate medium contains highly nutritious extract and the 5 % blood supplement which provides additional use in isolation, cultivation, and determination of hemolytic reactions of fastidious pathogenic microorganisms. Hemolytic patterns may vary with the source of animal blood or type of base medium used. In sheep blood, nucleotidase destroys V factors preventing the growth of *Haemophilus* species on sheep blood agar unless other microorganisms, such as Staphylococci, provide the V factors. Small amounts of reducing sugars inhibit the expression of β-hemolysis, and β-hemolytic Streptococci may develop a green zone or ring of hemolysis.

A number of streptococcal species produce substances that lyse the red cell wall releasing hemoglobin. Such substances are referred to as hemolysins. The activity of streptococcal hemolysins, streptolysins, can be readily observed when the organisms are growing on a blood agar plate. Different *Streptococci* produce different effects on the red blood cells in blood agar.

- Those that achieve incomplete hemolysis and only partial destruction of the cells around colonies are called alpha-hemolytic Streptococci. Characteristically, this type of hemolysis is seen as a distinct greening of the agar in the hemolytic zone, and thus this group of streptococci has also been referred to as the viridans group.
- Species whose hemolysins cause complete destruction of red cells in the agar zones surrounding their colonies are said to be beta-hemolytic and are represented as small opaque or semi translucent colonies surrounded by clear zones in a red opaque medium. Two types of beta lysins are produced; Streptolysin-O, an antigenic oxygen-labile enzyme and Streptolysin-S, a non-antigenic oxygen-stable enzyme. The hemolytic reaction is enhanced when blood agar plates are streaked and simultaneously stabbed to show subsurface hemolysis by Streptolysin-O in an environment with reduced oxygen tension. Some strains of *Staphylococcus*, *Escherichia coli*, and other bacteria also may show beta hemolysis.
- Some species of Streptococci do not produce hemolysins. Therefore, when their colonies grow on blood agar, no change is seen in the red blood cells around them. These species are referred to as nonhemolytic or gamma hemolytic Streptococci.

COMPOSITION

Ingredients	Gms / Ltr
Casein enzymic hydrolysate	14.000
Agar	12.500
Sodium chloride	5.000
Peptic digest of animal tissue	4.500
Yeast extract	4.500
Sheep Blood	50.000 ml

PRINCIPLE

Medium contains nutritional components like pancreatic digest of casein, neutralized peptone, and yeast extract, and the addition of sodium chloride provides an osmotically balanced medium for bacterial cells. The addition of 5% defibrinated sheep blood allows for the determination of hemolytic reactions, an important differential characteristic.

INSTRUCTION FOR USE

Either streak, inoculate or surface spread the test inoculum aseptically on the plate.

QUALITY CONTROL SPECIFICATIONS

Appearance	:	Cherry red colour, opaque gel.
Quantity of Medium	:	25ml of medium in 90mm plates.
pH (at 25°C)	:	7.3± 0.2
Sterility Check	:	Passes release criteria

INTERPRETATION

Cultural characteristics observed after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Haemolysis	Incubation Temperature	Incubation Period
Streptococcus pneumonia	6303	50-100	Luxuriant	>=70%	Alpha	35-37°C	18 - 48 Hours
Streptococcus pyogenes	19615	50-100	Luxuriant	>=70%	Beta	35-37°C	18 - 48 Hours
Staphylococcus aureus	6538	50-100	Luxuriant	>=70%	Beta	35-37°C	18 - 48 Hours

PACKAGING:

Doubled layered packing containing 5 No. of plates with one silica gel desiccant bag packed inside it.

STORAGE

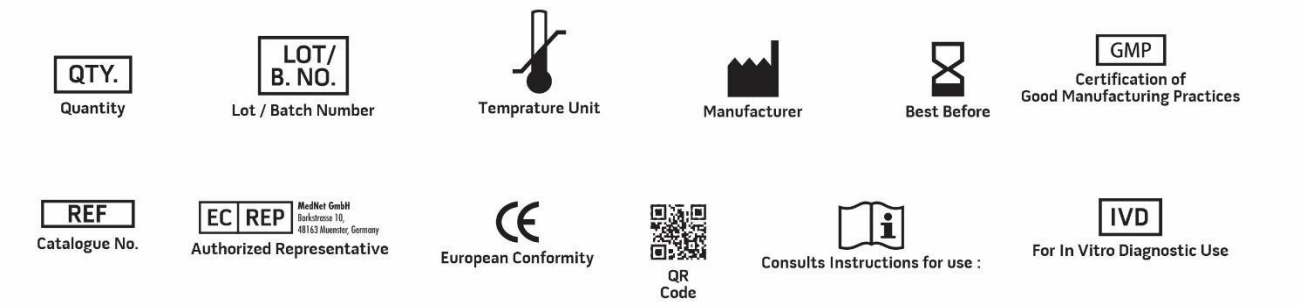
On receipt, store the plates at 2-8 °C. Avoid freezing and overheating. Do not open until ready to use. Prepared plates stored in their original sleeve wrapping until just prior to use may be inoculated up to the expiration date and incubated for recommended incubation times. Allow the medium to warm to room temperature before inoculation.
Product Deterioration: Do not use plates if they show evidence of microbial contamination, discoloration, drying, cracking or other signs of deterioration.

DISPOSAL

After use, prepared plates, specimen/sample containers and other contaminated materials must be sterilized before discarding.

REFERENCES

1. Pelczar M. J. Jr., Reid R. D., Chan E. C. S., 1977, Microbiology, 4th Ed., Tata McGraw-Hill Publishing Company Ltd, New Delhi.
2 .Koneman E. W., Allen S. D., Janda W. M., Schreckenberger P. C., Winn W. C. Jr., 1992, Colour Atlas and Textbook of Diagnostic Microbiology, 4th Ed., J. B. Lippinccott Company.
3. Spector W. S., (Ed.), 1961, Handbook of Biological Data, W. B. Saunder Company, Philadelphia and London.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.

***For Lab Use Only**

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