

TM 182 - M17 AGAR BASE

INTENDED USE

For cultivation of lactic Streptococci and plaque assay of lactic bacteriophages.

PRODUCT SUMMARY AND EXPLANATION

M17 media are based on the formulation described by Terzaghi and Sandine for the cultivation and enumeration of lactic Streptococci and their bacteriophages. It is possible to study plaque morphology and lysogeny. M17 Agar is recommended by the International Dairy Federation for selective enumeration of *Streptococcus thermophilus* from yoghurt. M17 Agar is recommended by APHA for the cultivation of lactic Streptococci. Lactic Streptococci are nutritionally fastidious and require complex media for optimal growth. Disodium glycerophosphate maintains the pH above 5. The maintenance of pH is very important as lower pH results in injury and reduced recovery of lactic Streptococci. Glycerophosphate does not form precipitate with calcium which is needed for the plaque assay of lactic bacteriophages.

M17 Agar is also recommended by the International Dairy Federation for selective enumeration of *Streptococcus thermophilus* from yoghurt. It is also suitable for cultivation and maintenance of starter cultures for cheese and yoghurt manufacturing. This medium helps in detecting streptococcal mutants that are lactose non-fermenters. Suggested technique to enumerate streptococci is to seed in mass or by stabbing with agar, melted and cooled to 50-55°C, and incubating them at 42°C for 24 hours' period. With these conditions, all the colonies might be streptococci. Longer incubation periods or lower temperatures may cause morphological changes in the colonies, which hinders in the recognition of the colonies. Lactose-positive colonies of streptococci are visible after 15 hours and after 5 days they may reach a diameter of about 3-4 mm, whereas those of lactose-negative are 1 mm in diameter. Bacteriophages presence is observed by appearance of characteristic plaques over the bacterial growth.

COMPOSITION

Ingredients	Gms / Ltr
Peptone	5.000
Soya peptone	5.000
Yeast extract	2.500
Beef extract	5.000
Ascorbic acid	0.500
Magnesium sulphate	0.250
Lactose	5.000
Agar	10.000

PRINCIPLE

Peptone, soya peptone, yeast extract and Beef extract provide carbonaceous, nitrogenous compounds, long chain amino acids, vitamin B complex and other essential growth factors. Lactose is the fermentable carbohydrate and ascorbic acid is stimulatory for the growth of lactic Streptococci. Magnesium sulphate provides essential ions to the organisms. Disodium-glycerophosphate maintains the pH above 5. The maintenance of pH is very important as lower pH results in injury and reduced recovery of lactic Streptococci. Disodium glycerophosphate suppresses *Lactobacillus bulgaricus*. Shankar and Davies reported isolation and enumeration of *Streptococcus thermophilus* from yoghurt.

INSTRUCTION FOR USE

- Dissolve 33.25 grams in 1000 ml purified/distilled water.
- Add 19 grams of Disodium β-Glycerophosphate.
- Heat to boiling to dissolve the medium completely.



- Sterilize by autoclaving at 15 psi pressure (121°C) for 15 minutes.
- Cool to 45-50°C. Mix well and dispense as desired.

QUALITY CONTROL SPECIFICATIONS

- Appearance of Powder** : Cream to yellow homogeneous free flowing powder
- Appearance of prepared medium** : Light yellow coloured clear to slightly opalescent gel forms in Petri plates
- pH (at 25°C)** : 7.1±0.2

INTERPRETATION

Cultural characteristics observed after an incubation with added Disodium β- Glycerophosphate.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Incubation Temperature	Incubation Period
<i>Enterococcus faecalis</i>	29212	50-100	good-luxuriant	≥50 %	35-37°C	24-48 Hours
<i>Lactobacillus bulgaricus subsp. bulgaricus</i>	11842	50-100	none-poor	0-10%	35-37°C	24-48 Hours
<i>Lactobacillus leichmannii</i>	4797	50-100	good-luxuriant	≥50 %	35-37°C	24-48 Hours
<i>Lactobacillus plantarum</i>	8014	50-100	good-luxuriant	≥50 %	35-37°C	24-48 Hours
<i>Streptococcus thermophilus</i>	14485	50-100	good-luxuriant	≥50 %	35-37°C	24-48 Hours

PACKAGING:

In pack size of 500 gm bottles.

STORAGE

Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided on the container. After the desired amount of medium has been taken out replace the cap tightly to protect from hydration.

Product Deterioration: Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

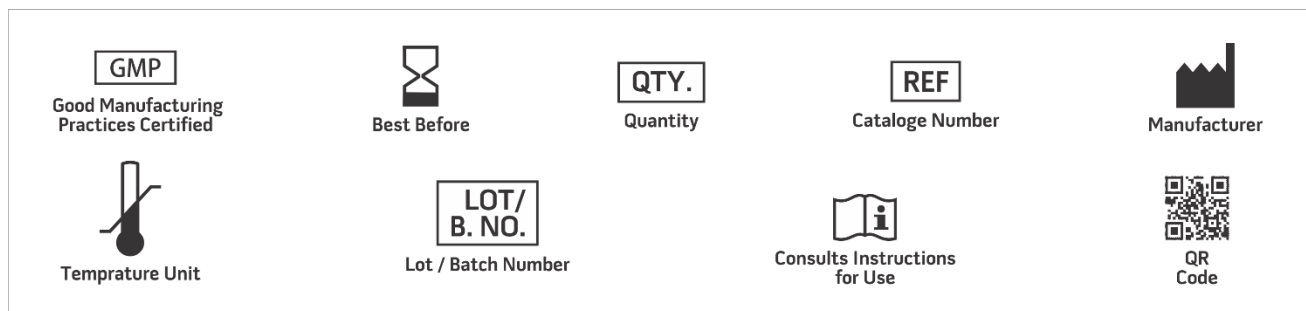
DISPOSAL

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



REFERENCES

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5. 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.
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8. Shankar P.A. and Davies F.L., 1977, Soc. Dairy Technol., 30:28.
9. Terzaghi B.E. and Sandine W.E., 1975, Appl. Microbiol., 29:807.
10. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed., APHA Inc., Washington, D.C.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices.
***For Lab Use Only**
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