TM 2154 – LEE'S AGAR

INTENDED USE

For differential enumeration of yoghurt starter bacteria (*Lactobacillus bulgaricus, Streptococcus thermophilus*).

PRODUCT SUMMARY AND EXPLANATION

Yoghurt is a fermented milk product in which *Streptococcus thermophilus* and *Lactobacillus bulgaricus* are the essential microbial species that are active in a symbiotic relationship. To obtain optimum consistency, flavour and odour, the two species should be present in about equal numbers in the culture. Dominance by either species can cause defects. Lees Agar, described by Lee et al is used for the differential enumeration of yoghurt starter bacteria. This medium is also recommended by APHA for the same purpose.

Lees Agar contains sucrose, which most *L. bulgaricus* strains will not ferment, but *S. thermophilus* will, and lactose, which both species utilize. With a suitable combination of sucrose and lactose, the rate of acid production by *S. thermophilus* is enhanced and that of *L. bulgaricus* restricted. Therefore, Streptococci grow first and produce a creamy, buttery aroma from diacetyl and similar metabolites. The redox potential is also thus, lowered by Streptococci, which enables Lactobacilli to grow, thereby growth stimulatory products for Streptococci are synthesized by Lactobacilli. Hence the typical sharp acetaldehyde flavour of mature yoghurt is formed.

COMPOSITION

Ingredients	Gms / Ltr
Tryptone	10.000
Yeast extract	10.000
Lactose	5.000
Sucrose	5.000
Calcium carbonate	3.000
Dipotassium hydrogen phosphate	0.500
Bromocresol purple	0.020
Agar	18.000

PRINCIPLE

This medium consists of Tryptone and yeast extract which provide the essential nitrogenous nutrients to the yoghurt (lactic) starter bacteria. Lactose and sucrose are the fermentable carbohydrates. Calcium carbonate along with dipotassium phosphate is added to buffer the medium and avoid the drastic drop in pH due to lactic acid formation. Bromocresol purple is the pH indicator, which turns yellow in acidic condition and imparts yellow colour to the colony. It is recommended to dry the media plates for 18-24 hours prior to use.

INSTRUCTION FOR USE

- Dissolve 51.52 grams in 1000 ml purified/distilled water.
- Heat just to boiling and sterilize by autoclaving at 15 psi pressure (121°C) for 20 minutes.
- Cool to 45-50°C. While dispensing, mix carefully to suspend calcium carbonate evenly. Pour into sterile Petri plates to obtain 4-5 mm thick gel.
  Note: Due to the presence of calcium carbonate, the prepared medium forms opalescent solution with white precipitate.

QUALITY CONTROL SPECIFICATIONS

Appearance of Powder : Light yellow to light grey homogeneous free flowing powder.
Appearance of prepared medium : Purple coloured, opaque gel forms in Petri plates.
pH (at 25°C) : 7.0 ± 0.2

INTERPRETATION

Cultural characteristics observed in presence of Carbon dioxide after incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Colour of colony	Incubation Temperature	Incubation Period
Lactobacillus bulgaricus	11842	50-100	Luxuriant	>=70%	White	35-37°C	48 Hours
Streptococcus thermophilus	14485	50-100	Luxuriant	>=70%	Yellow	35-37°C	48 Hours

PACKAGING:

In pack size of 500 gm bottles.

STORAGE

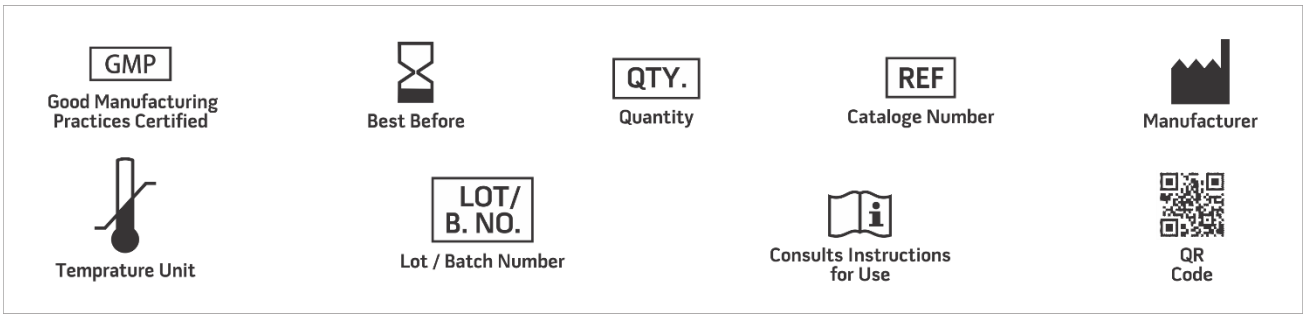
Dehydrated powder, hygroscopic in nature, store in a dry place, in tightly-sealed containers between 25-30°C and protect from direct sunlight. Under optimal conditions, the medium has a shelf life of 4 years. When the container is opened for the first time, note the time and date on the label space provided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

1. Davis J. G., Ashton T. F. and MaCaskill M., 1971, Dairy Ind., 36:569.
2. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
3. 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.
4. Lee S. Y., Vedamuthu E. R., Washam C. J. and Reinbold G. W., 1974, J . Milk Food Technol., 37: 272.



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

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