

# TM 1351 - HYA AGAR

## **INTENDED USE**

For differentiation of *Lactobacillus bulgaricus* and *Streptococcus thermophilis* on the basis of colony morphology from yoghurt cultures.

## **PRODUCT SUMMARY AND EXPLANATION**

Yoghurt is a fermentable milk product in which *Streptococcus thermophilus* and *Lactobacillus bulgaricus* are the essential microbial species and are active in a symbiotic relationship. The large number of media proposed for lactic acid bacteria, particularly for Streptococci and lactobacilli is indicative of the difficulties encountered in growing some strains of these organisms. The choice of medium is governed to some extent by the particular strains under study and therefore by products or habitat. In general, lactic acid bacteria are tolerant to low pH, they can be very sensitive to other adverse conditions. Samples to be examined for numbers of viable lactic acid bacteria should not be frozen prior to analysis. Porubcan and Sellars described this medium on which *L. bulgaricus* grow as diffuse, low mass colonies (2-10 mm in diameter) and *S. thermophilus* as discrete high mass colonies (1-3 mm in diameter). To obtain optimum consistency, flavour and odour, many investigators claim that the two species should be present in about equal numbers in the culture. Dominance by either species can cause defects. Because of the emphasis on maintaining balance between coccus and rods, techniques are needed to determine the relative proportion of *S. thermophilus* and *L.bulgaricus* when grown together in milk culture. Differentiation of two species on HYA Agar is based on colony morphology. Also this media is recommended by APHA.

# COMPOSITION

Ingredients	Gms / Ltr	
Beef extract	1.000	
Proteose peptone	10.000	
Dextrose (Glucose)	2.500	
Galactose	2.500	
Lactose	5.000	
Agar	15.000	

# PRINCIPLE

Beef extract and proteose peptone provides necessary nitrogenous nutrients required for growth of two species. The sugars dextrose, galactose, lactose serves as energy sources.

### **INSTRUCTION FOR USE**

- Dissolve 36.0 grams in 1000 ml purified/distilled water.
- Heat to boiling to dissolve the medium completely.
- Sterilize by autoclaving at 15 psi pressure (121°C) for 20 minutes.
- Cool to 45-50°C. Mix well and pour into sterile Petri plates.

# QUALITY CONTROL SPECIFICATIONS

Appearance of Powder	: Cream to yellow homogeneous free flowing powder.		
Appearance of prepared medium	: Yellow coloured, clear to slightly opalescent gel forms in Petri plates.		
pH (at 25°C)	: 6.8±0.2		

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## INTERPRETATION

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### Cultural characteristics observed after an incubation.

Microorganism	ATCC	Inoculum (CFU/ml)	Growth	Recovery	Incubation Temperature	Incubation Period
Lactobacillus bulgaricus	11842	50-100	Luxuriant	>=70%	35-37°C	24-48 Hours
Streptococcus thermophilus	14485	50-100	Luxuriant	>=70%	35-37°C	24-48 Hours

# PACKAGING:

In pack size of 100 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.

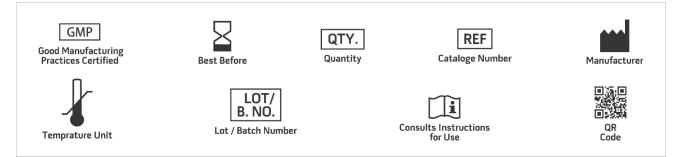
**Product Deterioration:** Do not use if they show evidence of microbial contamination, discoloration, drying or any other signs of deterioration.

## DISPOSAL

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

#### REFERENCES

- 1. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
- 2. 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.
- 3. Porubcan R. S., and Sellars R. L., 1973, J. Dairy Sci., 56: 634.
- 4. Salfinger Y., and Tortorello M.L. Fifth (Ed.), 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.



NOTE: Please consult the Material Safety Data Sheet for information regarding hazards and safe handling Practices. \*For Lab Use Only Revision: 08 Nov., 2019

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