

Technical Information

Modified Rogosa Agar (M16 Agar)

Product Code: DM 1600

Application: M16 Agar is recommended for cultivation and enumeration of lactic streptococci used in manufacture of cheddar cheese.

Composition**

Ingredients	Gms / Litre			
Papaic digest of soyabean meal	5.000			
Tryptose	5.000			
Beef extract Yeast extract	5.000 2.500			
Dextrose Ascorbic acid	5.000 0.500			
Sodium acetate Agar	3.000 10.000			
Final pH (at 25°C) **Formula adjusted, standardized to suit perfor	7.2±0.2			

Principle & Interpretation

One of the most important groups of acid producing bacteria in food industry is the lactic acid bacteria. There acid-producing bacteria are found in nature, in the soil, on raw agricultural products and in certain processed foods. Streptococci belong to the lactic acid bacteria group is a genus of spherical, gram-positive bacteria, and a member of the phylum Firmicutes (1).

M16 Agar is a modification of Rogosa Sodium Lactate Agar recommended by APHA ^(1, 3). This medium was developed to support growth of lactic streptococci used in cheddar cheese manufacturing in New Zealand ⁽²⁾. This medium can also be used as selective medium for the cultivation of oral and faecal lactobacilli. Since some lactobacilli do not grow on this medium if incubated aerobically therefore incubation in CO₂-enriched atmosphere is recommended for their growth.

The large number of media proposed for lactic acid bacteria, particularly for streptococci and /or lactobacilli, is an indicative of the variability in growth features of different species, thereby the difficulties encountered in growing some strains of this group of organisms. While the lactic acid bacteria in general are acid tolerant at low pH, but can be very sensitive to other adverse conditions. Freezing and thawing prior to analysis may be detrimental to cell growth. Dilution process may also damage lactic acid bacteria in samples for which sterile 0.1% Peptone Water (DM1028) in used as the diluent.

Papaic digest of soyabean meal, tryptose and beef extract provide the essential nutrients like amino acids, minerals etc. Yeast extracts supplies vitamin B complex to the lactic streptococci. Dextrose is the fermentable carbohydrate and energy source. Sodium acetate inhibits other contaminating bacteria and suppresses swarming growth. Ascorbic acid provides vitamin C to the organisms.

The samples to be tested are processed to enumerate bacteria by pour plate technique.

Methodology

Suspend 36 grams of powder media in 1000 ml distilled water. Shake well & heat to boiling to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Mix well and pour into sterile Petri plates.





Quality Control

Physical Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.0% Agar gel.

Colour and Clarity of prepared medium

Light amber coloured clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 3.6% w/v aqueous solution at 25°C. pH: 7.2±0.2

pH Range

7.00-7.40

Cultural Response/Characteristics

DM 1600: Cultural characteristics observed after an incubation at 35-37 $^\circ$ C for 18-48 hours in CO $_2$ enriched atmosphere.

Organism	Inoculum (CFU)	Growth	Recovery
Lactobacillus lactis ATCC 8000	50-100	good-luxuriant	>=50%
Streptococcus cremoris ATCC 19257	50-100	good-luxuriant	>=50%

Storage and Shelf Life

Dried Media: Store below 30°C in tightly closed container and use before expiry date as mentioned on the label. **Prepared Media:** 2-8⁰ in sealable plastic bags for 2-5 days.

Further Reading

- 1. Downes F. P. and Ito K., (Eds.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., APHA, Washington, D.C.
- 2. Lowrie R. J. and Pearce L. E., 1971, New Zealand, J. Dairy Sci. Technol., 6: 166.
- 3. Rogosa M., Mitchell J. A. and Wiseman R. F., 1951, J. Bacteriol., 62: 132-133

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