



Litmus Milk

M609S

Litmus Milk is recommended for maintenance of *Lactobacilli* and for determining the action of bacteria on milk. It is recommended by BIS committee under specifications IS:5887(Part-IV)-1976.

Composition**

Ingredients	Gms / Litre
Skim milk powder	100.000
Litmus	5.000
Final pH (at 25°C)	6.8±0.1

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 105 grams in 1000 ml distilled water, agitating continuously. Dispense 10 ml amounts into 15 x 150 mm. tubes and sterilize by autoclaving at 15 lbs pressure (121°C) for 5 minutes. AVOID OVERHEATING.

Principle And Interpretation

Litmus Milk is the most useful medium in dairy industry as it is a reliable indicator of bacterial action on milk (1). Litmus is a good indicator of acidity, alkalinity and its oxidation-reduction potential is useful in milk media with lower toxicity to microorganisms than bromo cresol purple (2). Addition of dextrose 1% w/v and/or yeast extract 5% w/v to Litmus Milk accelerates the growth of some organisms which cannot grow in plain Litmus Milk (1,2,3). Present formulation is recommended by BIS (4) for detection of bacteria responsible for food poisoning.

For detection of *Clostridium perfringens* in water, inoculate freshly heated tubes of Litmus Milk with various quantities of water and heat at 80°C for 10-15 minutes to destroy nonsporing organisms. Examine after every 24 hours for positive Stormy Clot reaction at 35°C up to 5 days (5). Anaerobiosis in Litmus Milk can be obtained by adding a small heated iron nail or 0.1 gram of reduced iron to the medium (6). Skim milk is the substrate, metabolized by particular species of bacteria in different ways. The actions of bacteria can be categorized as follows:

ACID REACTION

1. Pink to red colour of milk - Fermentation of lactose and/or dextrose in milk.
2. Acid coagulation - Lactic acid production, producing a casein curd in clear watery fluid.
3. Stormy clot - Gas formation in coagulated casein curd.

ALKALINE REACTION

1. Blue colour of the milk - Formation of basic amines or ammonia due to proteolysis.
2. Alkaline coagulation with a soft blue clot - Paracasein formation from casein by enzyme rennin.
3. Peptonization - Digestion of casein, evident by clearing of the medium and dissolution of the clot.

REDOX REACTION

1. Decolourized medium (Similar to freshly autoclaved Litmus Milk) - Reaction of Litmus in the depths of the tube by reductase enzymes with the resultant removal of oxygen to form the decolourized leucolitmus compound.

Reactions obtained in this medium are not specific and further tests must be carried out.

Quality Control

Appearance

Pinkish purple to grey coloured Homogeneous free flowing powder may have small particles

Colour and Clarity of prepared medium

Light purple coloured opaque, milky solution

Reaction

Reaction of 10.5% w/v aqueous solution at 25°C. pH : 6.8±0.1

pH

6.70-6.90

Cultural Response

Cultural characteristics observed after an incubation 35-37°C for upto 14 days and record the reactions of various intervals during the incubation.

Cultural Response

Organism	Growth	Reaction
<i>Bacillus cereus</i> ATCC 10876	good-luxuriant	peptonization with or without coagulation
<i>Clostridium perfringens</i> ATCC 13124	good-luxuriant	stormy fermentation (Gas)
<i>Lactobacillus acidophilus</i> ATCC 11506	good-luxuriant	acid clot (pink)
<i>Pseudomonas aeruginosa</i> ATCC 27853	good-luxuriant	peptonization (Clearing)

Storage and Shelf Life

Store below 30°C in tightly closed container and the prepared medium at 2 - 8°C. Use before expiry date on the label.

Reference

1. Davis J. G., 1935, J. Dairy Res., 6:121
2. Davis J. G., 1955, A Dictionary of Dairying, 2nd ed, Leonard Hill.
3. Davis J. G., 1959, Milk Testing, 2nd ed., United Trade Press.
4. Bureau of Indian Standards IS : 5887 (Part IV) 1976.
5. Department of Health and Social security, 1969, Report No. 21, HMSO, London.
6. Townsend C.T., Somers J. J., Lamb F. C. and Olson N. A., 1956, A Laboratory Manual for the Canning Industry, 2nd ed., National Canners Association, Washington.
7. MacFaddin J. F., 1985, Media for Isolation-Cultivation-Maintenance of Medical Bacteria, Vol. I, Williams and Wilkins, Baltimore.

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