



Lactobacilli Broth, AOAC

M367

Lactobacilli Broth, AOAC, is used for preparation of inocula of test bacteria used in microbiological assays of the B-vitamins.

Composition**

Ingredients	Gms / Litre
Peptonized milk	15.000
Yeast extract	5.000
Dextrose	10.000
Tomato juice (100 ml)	5.000
Monopotassium phosphate	2.000
Polysorbate 80	1.000
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 38 grams in 1000 ml distilled water. Heat, if necessary, to boiling to dissolve the medium completely. Distribute into tubes in 10 ml amounts and sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes.

Principle And Interpretation

Lactobacilli Broth, AOAC was formulated by Loy (1) and recommended by AOAC (2) for preparing inocula of test bacteria used for Microbiological assay of Vitamin B. Stock cultures of *Lactobacillus leichmanni* ATCC 7830, *Lactobacillus plantarum* ATCC 8014, *Lactobacillus casei* ATCC 7469, *Enterococcus hirae* ATCC 8043 and other such B vitamin requiring strains are prepared by stab inoculation of sterile Lactobacillus Agar, AOAC and incubated for 18-24 hours at a constant temperature between 30-40°C. Lactobacilli Broth, AOAC is used for cultivation and preparation of inocula of the above mentioned stock cultures (3). Inoculum is prepared by inoculating these cultures in Lactobacillus Broth, AOAC and incubating at 35-37°C.

Peptonized milk and yeast extract provide essential growth nutrients. Dextrose is the energy source. Phosphate provides buffering system while tomato juice helps in lowering the pH. Polysorbate 80 supplies fatty acids.

Before using a culture in any assay, at least 2 successive transfers during a 1-2 week period are essential. Any culture older than one week should not be used.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Medium amber coloured clear solution in tubes

Reaction

Reaction of 3.8% w/v aqueous solution at 25°C. pH : 6.8±0.2

pH

6.60-7.00

Cultural Response

M367: Cultural characteristics observed after an incubation at 35 - 37°C for 18 - 48 hours

Organism	Inoculum (CFU)	Growth
<i>Enterococcus hirae</i> ATCC 8043	50-100	luxuriant

<i>Lactobacillus casei</i> ATCC 7469	50-100	luxuriant
<i>Lactobacillus leichmannii</i> ATCC 7830	50-100	luxuriant
<i>Lactobacillus plantarum</i> ATCC 8014	50-100	luxuriant

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. Loy, 1958, J. AOAC, 4:61.
2. Williams, (Ed.), 2005, Official Methods of Analysis of the Association of Official Analytical Chemists, 19th Ed., AOAC, Washington, D.C.
3. Atlas R. M., 2004, Handbook of Microbiological Media, 3rd Edition, CRC Press

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