



Technical Data

Lactobacillus Selection Bile Agar Base (LBS Bile Agar Base)

M1165

Intended Use:

Recommended for the selective isolation, cultivation and enumeration of Lactobacilli.

Composition**

Ingredients	g / L
Tryptone	10.000
Yeast extract	5.000
Dextrose (Glucose)	20.000
Sodium acetate	25.000
Potassium dihydrogen phosphate	6.000
Ammonium citrate	2.000
Bile □	1.500
Polysorbate 80 (Tween 80)	1.000
Magnesium sulphate	0.575
Manganese sulphate	0.120
Ferrous sulphate	0.034
Agar	15.000
Final pH (at 25°C)	5.4±0.2

**Formula adjusted, standardized to suit performance parameters

□Equivalent to Oxgall

Directions

Suspend 86.23 grams in 1000 ml purified/distilled water containing 1.32 ml glacial acetic acid. Heat to boiling with stirring to dissolve the medium completely. Boil for 2-3 minutes. DO NOT AUTOCLAVE. If storage is necessary, autoclave at $\Delta 118^{\circ}\text{C}$ for 15 minutes. Cool to $45-50^{\circ}\text{C}$. Mix well and pour into sterile Petri plates.

Δ corresponds to 12 lbs pressure.

Principle And Interpretation

Lactobacilli grow in a variety of habitats, wherever high levels of soluble carbohydrate, protein background products, vitamins and a low oxygen tension occur (1). These sites include the oral cavity, the intestinal tract (2,3), the vagina (4), food products (5) and dairy products (6). LBS Bile Agar Base, formulated by Gillil and and Speck (7) is recommended by APHA for the isolation and enumeration of lactobacilli. LBS Bile Agar Base is similar in composition to Lactobacillus Selection Agar Base, the only difference being the additional bile added to the former .

Tryptone and yeast extract serve as sources of essential nutrients. Dextrose is the carbohydrate and energy source. Polysorbate 80 serves as an additional source of growth factors and fatty acids required for metabolism of *Lactobacillus* species. Selectivity of the medium is obtained due to the presence of ammonium citrate and sodium acetate. These inhibit the accompanying microbial and fungal flora and also restrict swarming of colonies (8). The low acidic pH of the medium obtained by addition of glacial acetic acid is inhibitory to several bacterial species. Sulphates provide essential ions. LBS Bile Agar Base is made selective for bile-resistant lactobacilli by incorporating 0.15% bile.

Type of specimen

Food and dairy samples

Specimen Collection and Handling:

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (5,6).

After use, contaminated materials must be sterilized by autoclaving before discarding.

Warning and Precautions :

Read the label before opening the container. Wear protective gloves/protective clothing/eye protection/face protection. Follow good microbiological lab practices while handling specimens and culture. Standard precautions as per established guidelines should be followed while handling specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. Do not overheat; prolonged heating causes medium to darken and some types of lactobacilli present in the small numbers may not develop to full size, appear as pinpoint colonies.
2. Further serological and biochemical testing is required for complete identification.

Performance and Evaluation

Performance of the medium is expected when used as per the direction on the label within the expiry period when stored at recommended temperature.

Quality Control

Appearance

Cream to yellow homogeneous free flowing powder

Gelling

Firm, comparable with 1.5% Agar gel

Colour and Clarity of prepared medium

Yellow coloured clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of 8.62% w/v aqueous solution at 25°C. pH : 5.4±0.2

pH

5.20-5.60

Cultural Response

Cultural characteristics observed in presence of 3-5% Carbon dioxide(CO₂) after an incubation at 35-37°C for 48 hours.

Organism	Inoculum (CFU)	Growth	Recovery
<i>Lactobacillus acidophilus</i> ATCC 4356	50-100	luxuriant	≥50%
<i>Lactiplantibacillus plantarum</i> ATCC 8014	50-100	luxuriant	≥50%
<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 ⁴	inhibited	0%
<i>Staphylococcus aureus</i> subsp. <i>aureus</i> ATCC 25923 (00034*)	≥10 ⁴	inhibited	0%
<i>Lactobacillus rhamnosus</i> ATCC 9595	50-100	luxuriant	≥50%
<i>Enterococcus faecalis</i> ATCC 29212 (00087*)	≥10 ⁴	inhibited	0%
## <i>Proteus hauseri</i> ATCC 13315	≥10 ⁴	inhibited	0%

Key: * Corresponding WDCM numbers.

Formerly known as *Proteus vulgaris*

\$ Formerly known as *Lactobacillus plantarum*

Storage and Shelf Life

Store dehydrated powder and the prepared medium at 2-8°C. Use before expiry date on the label. On opening, product should be properly stored dry, after tightly capping the bottle in order to prevent lump formation due to the hygroscopic nature of the product. Improper storage of the product may lead to lump formation. Store in dry ventilated area protected from extremes of temperature and sources of ignition Seal the container tightly after use. Product performance is best if used within stated expiry period.

Disposal

User must ensure safe disposal by autoclaving and/or incineration of used or unusable preparations of this product. Follow established laboratory procedures in disposing of infectious materials and material that comes into contact with sample must be decontaminated and disposed of in accordance with current laboratory techniques (9,10).

Reference

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5. 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.
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