



Technical Data

Lactic Acid Bacteria Selective Broth Base (Raka Ray No. 3 Broth Base)

M1384

Intended Use:

Recommended for selective isolation of lactic acid bacteria encountered in beer and the brewing process.

Composition**

Ingredients	Gms / Litre
Tryptone	20.000
Yeast extract	5.000
HL extract#	1.000
Maltose	10.000
Fructose	5.000
Dextrose (Glucose)	5.000
Betaine hydrochloride	2.000
Diammonium hydrogen citrate	2.000
L-Aspartic acid	2.500
Magnesium sulphate	0.980
Manganese sulphate	0.420
Dipotassium hydrogen phosphate	2.000
N-acetyl glucosamine	0.500
Potassium glutamate	2.500
Final pH (at 25°C)	5.4±0.2

**Formula adjusted, standardized to suit performance parameters

Equivalent to Liver extract

Directions

Suspend 29.45 grams in 500 ml purified/distilled water containing 5 ml Polysorbate 80. Heat if necessary to dissolve the medium completely. Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. Cool to 45-50°C and aseptically add rehydrated contents of 1 vial of Lactic Supplement (FD055). Mix well and dispense into sterile tubes or flasks as desired.

Principle And Interpretation

Lactic Acid Bacteria Selective Medium was formulated by Saha, Sondag and Middlekauff to monitor the brewing process and analyze it for a wide range of bacteria (10). These media are also recommended by the American Society of Brewing Chemists (ASBC) and the European Brewing Convention (EBC) (1,8). Lactic Acid Bacteria Selective Medium was found to be superior to several other media for the cultivation of Lactobacilli and Pediococci (2,3,11). Lactic Acid Bacteria Selective Broth Base also suppressed the growth of non-lactic acid facultative bacteria that are often associated with lactic beer spoilage (9).

Yeast extract, tryptone and HL extract serve as sources of carbon, nitrogen, vitamins, amino acids and essential nutrients. Dextrose (glucose), maltose and fructose serve as sources of carbon and energy. Fructose is an essential carbohydrate for the growth for *Lactobacillus fructivorans* (11). Maltose helps to detect glucose non-fermenting lactobacilli (6). Polysorbate 80, maltose, yeast extract and N-acetyl glucosamine stimulates growth of lactobacilli (7). Various salts provide trace elements. Cycloheximide and phenyl ethanol (as FD) serves to inhibit yeast and gram-negative organisms respectively.

Type of specimen

Brewery samples

Specimen Collection and Handling

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. Further biochemical and serological tests must be carried out 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 beige homogeneous free flowing powder

Colour and Clarity of prepared medium

Dark amber coloured clear solution in tubes.

Reaction

Reaction of 5.89% w/v aqueous solution at 25°C. pH : 5.4±0.2

pH

5.20-5.60

Cultural Response

Cultural characteristics observed under anaerobic condition, with added Lactic Supplement (FD055), after an incubation at 25-30°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth
<i>Lactobacillus acidophilus</i> ATCC 11506	50-100	good-luxuriant
<i>Lactobacillus plantarum</i> ATCC 8014	50-100	good-luxuriant
<i>Lactobacillus fermentans</i> ATCC 9338	50-100	good-luxuriant
<i>Lactobacillus brevis</i> ATCC 367	50-100	good-luxuriant
<i>Lactobacillus buchneri</i> ATCC 11307	50-100	good-luxuriant
<i>Pedococcus acidilactis</i> ATCC 8042	50-100	good-luxuriant
<i>Escherichia coli</i> ATCC 25922 (00013*)	≥10 ⁴	inhibited
<i>Saccharomyces cerevisiae</i> ATCC 9763 (00058*)	≥10 ⁴	inhibited

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container 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 (4,5).

Reference

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10. Saha R. B., Sondag R. J. and Middlekauff J. E., 1974, An improved medium for the selective culturing of lactic acid bacteria, Proceedings of the American Society of Brewing Chemists, 9th Congress, p. 9-10.
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Revision :02/2020

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