Lactobacillus Bulgaricus Agar Base

M927

Intended Use:

With acetate buffer is used for isolation and identification of *Lactobacillus bulgaricus* from clinical and non-clinical samples.

Composition**

Ingredients	g / L
Tryptone	10.000
Yeast extract	5.000
HM peptone B #	10.000
Dextrose (Glucose)	20.000
Dipotassium hydrogen phosphate	2.000
Tomato juice	2.000
Polysorbate 80 (Tween 80)	1.000
Agar	20.000
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters
Equivalent to Beef extract

Directions

Suspend 70 grams in 920 ml purified/distilled water and heat to boiling to dissolve the medium completely. Add 80 ml Acetate Buffer (11.355% Sodium acetate and 0.99% Acetic acid). Sterilize by autoclaving at 15 lbs pressure (121°C) for 15 minutes. **DO NOT OVERHEAT THE MEDIUM.** Cool to 45-50°C. Mix well and pour into sterile Petri plates.

Principle And Interpretation

Lactobacillus bulgaricus (*Lactobacillus delbrueckii* subsp. *bulgaricus*) is one of several bacteria used for the production of Kisselo mlyako (Bulgarian) - "Sour milk" yoghurt (yogurt). The bacterium was first identified in 1905 by the Bulgarian doctor Stamen Grigorov. It is named after Bulgaria, the country where it was first used (it thrives freely on the Balkan Peninsula). The bacterium feeds on milk and produces lactic acid which also helps to preserve the milk. Lactobacillus Bulgaricus Agar was originally formulated by Kulp and White (1) for the recovery of Lactobacilli. Further modification is recommended by APHA (2) for isolation and identification of *L. bulgaricus* from foods. *Streptococcus thermophilus* and *L. bulgaricus* are the essential microbial species and are active in symbiotic relationship in yoghurt. Because of the emphasis on maintaining a balance between cocci and rods, techniques are needed to determine the relative proportions of *S. thermophilus* and *L. bulgaricus* when grown together in milk cultures.

Tryptone, yeast extract and HM peptone B in the medium provide nitrogenous compounds, minerals, vitamins and trace ingredients. Polysorbate 80 supplies fatty acids required for the metabolism of Lactobacilli. Dextrose is the fermentable carbohydrate. Tomato juice along with acetate maintains the low pH of the medium and thus inhibits microorganisms other than Lactobacilli. Acetate also restricts the swarming of L. bulgaricus and along with dipotassium phosphate forms the buffering system.

Type of specimen

Clinical sample; Dairy sample- Yoghurt sample

Specimen Collection and Handling:

For clinical samples follow appropriate techniques for handling specimens as per established guidelines (3,4). For 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 :

In Vitro diagnostic use. For professional use only. 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 clinical specimens. Safety guidelines may be referred in individual safety data sheets.

Limitations :

1. **DO NOT OVERHEAT THE MEDIUM.**
2. Some strains may show poor growth due to nutritional variations.

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 2.0% agar gel.

Colour and Clarity of prepared medium

Medium amber coloured, clear to slightly opalescent gel forms in Petri plates

Reaction

Reaction of the medium (7% w/v aqueous solution of base containing 8%v/v acetate buffer) at 25°C. pH : 6.8±0.2

pH

6.60-7.00

Cultural Response

Cultural characteristics observed with added acetate buffer, after an incubation at 35-37°C for 18-48 hours.

Organism	Inoculum (CFU)	Growth	Recovery
Lactobacillus delbrueckii subsp. *bulgaricus* ATCC 11842 (00102*)	50-100	good-luxuriant	>=50%

Key : (*) Corresponding WDCM numbers.

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 20-30°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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (3,4).

Reference

1. Kulp W. L. and White V., 1932, Science, 76:17.
2. Salfinger Y., and Tortorello M.L., 2015, Compendium of Methods for the Microbiological Examination of Foods, 5th Ed., American Public Health Association, Washington, D.C.
3. Isenberg, H.D. Clinical Microbiology Procedures Handbook 2nd Edition.
4. Jorgensen, J.H., Pfaller, M.A., Carroll, K.C., Funke, G., Landry, M.L., Richter, S.S and Warnock., D.W. (2015) Manual of Clinical Microbiology, 11th Edition. Vol. 1.
5. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
6. Wehr H. M. and Frank J. H., 2004, Standard Methods for the Microbiological Examination of Dairy Products, 17th Ed.,APHA Inc., Washington, D.C.

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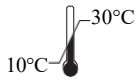
IVD ***In vitro*** **diagnostic medical device**



Storage temperature

EC REP CEpartner4U, Esdoornlaan 13,
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www.cepartner4u.eu

CE **CE Marking**

Do not use if package is damaged

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