



Arginine Dihydrolase Broth

Intended use

Recommended for detection of arginine dihydrolase producing microorganisms.

Composition**

Ingredients	Gms / Litre			
Peptone	1.000			
Sodium chloride	5.000			
Dipotassium hydrogen phosphate	0.300			
L-Arginine	10.000			
Bromo cresol purple	0.016			
Agar	3.000			
Final pH (at 25°C)	6.0±0.2			
**Formula adjusted, standardized to suit performance parameters				

Directions

Suspend 19.31 grams in 1000 ml purified / distilled water. Heat if necessary to dissolve the medium completely and distribute in 13x100 mm tubes. Sterilize by autoclaving at 115°C for 15 minutes. Allow the tubes to cool in an upright position.

Principle And Interpretation

Decarboxylase Media used for the detection of arginine dihydrolase and lysine and ornithine decarboxylase was first introduced by Moeller (6,7,8). Arginine Dihydrolase Broth is used for detection of arginine dihydrolase producing microorganisms. These types of media are used to differentiate bacteria on the basis of their decarboxylating activity towards the amino acids. Arginine decarboxylase enzyme is also known as Arginine dihydrolase. Arginine decarboxylase (or dihydrolase) production by various members of enteric bacteria aids in differentiating bacteria with closely related physiological characteristics (3). Bacteria producing arginine dihydrolase enzyme decarboxylate arginine present in this medium to putrescine. The production of amine, putrescine, elevates the pH. Bromo cresol purple is the pH indicator which forms purple colour in alkaline condition. Colour change from purple to yellow and then back to purple is positive reaction. Peptone provide the necessary nutrients to the organisms while L-arginine stimulates the arginine dihydrolase synthesis.

Dipotassium phosphate buffers the medium while sodium chloride maintains the osmotic balance. In differentiation of *Enterobacteriaceae*, control tubes without arginine must be used. If the tubes give positive purple reaction the test is considered as negative.

Type of specimen

Food and dairy samples; Water samples

Specimen Collection and Handling:

For food and dairy samples, follow appropriate techniques for sample collection and processing as per guidelines (1,9,10). For water samples, follow appropriate techniques for sample collection, processing as per guidelines and local standards. (2) 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. This medium is general purpose medium and may not support the growth of fastidious organisms.

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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

Light yellow to grey homogeneous free flowing powder

Gelling

Semisolid, comparable with 0.3% Agar gel.

Colour and Clarity of prepared medium

Purple coloured clear to slightly opalescent gel forms in tubes as butts

Reaction

Reaction of 1.93% w/v aqueous solution at 25°C. pH : 6.0±0.2

pН

5.80-6.20

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 18-24 hours.

Organism	Inoculum (CFU)	Growth	Motility	Arginine dihydrolase
#Klebsiella aerogenes ATCC 13048 (00175*)	50-100	luxuriant	positive,growth away from stabline causing turbidity	reaction, yellow
Klebsiella pneumoniae ATCC 13883 (00097*)	50-100	luxuriant	negative,growt along the stabline, surrounding medium remains clear	hnegative reaction, yellow colour or no colour change
Proteus vulgaris ATCC 13315	50-100	luxuriant	positive, growth away from stabline causing turbidity	negative reaction, yellow colour or no colour change
Salmonella Typhi ATCC 6539	50-100	luxuriant	positive,growth away from stabline causing turbidity	reaction, purple
<i>Salmonella</i> Typhimurium <i>ATCC 14028</i> (00031*)	50-100	luxuriant	positive,growth away from stabline causing turbidity	reaction, purple
Enterobacter sakazakii ATCC 12868	50-100	luxuriant	positive,growth away from stabline causing turbidity	reaction, purple

Key: *Corresponding WDCM numbers.

#- Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store between 10-30°C in a tightly closed container and the prepared medium at 15-25°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

- 1. American Public Health Association, Standard Methods for the Examination of Dairy Products, 1978, 14th Ed., Washington D.C.
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5. 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.

- 6. Moeller, 1954, Acta Pathol. Microbiol. Scand., 34:102.
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9.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.

 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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