



Moeller Decarboxylase Broth with Ornithine HCl

M688

Moeller Decarboxylase Broth with Ornithine hydrochloride is used to differentiate bacteria on the basis of their ability to decarboxylate L-Ornithine hydrochloride.

Composition**

Ingredients	Gms / Litre
Peptic digest of animal tissue	5.000
Beef extract	5.000
Dextrose	0.500
Bromocresol purple	0.010
Cresol red	0.005
Pyridoxal	0.005
L-Ornithine hydrochloride	10.000
Final pH (at 25°C)	6.0±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 20.52 grams in 1000 ml distilled water. Heat if necessary to dissolve the medium completely. Dispense in 5 ml amounts in screw capped tubes and sterilize by autoclaving at 15lbs pressure (121°C) for 10 minutes. Cool the tubes in an upright position. Inoculate the tubes and overlay with 2-3 ml of sterile mineral oil.

Principle And Interpretation

Moeller Decarboxylase Broth with Ornithine hydrochloride is used for differentiating gram-negative enteric bacilli on the basis of their ability to decarboxylate L-Ornithine hydrochloride. Decarboxylase Broth was introduced by Moeller for detecting the production of lysine and ornithine decarboxylase and arginine dihydrolase (1). Prior to Moellers work, bacterial amino acid decarboxylases were studied by Gale (2) and Gale and Epps (3). Production of ornithine decarboxylase is helpful criterion in differentiating *Klebsiella* and *Enterobacter* species. *Klebsiella* are nonmotile and do not produce ornithine decarboxylase while *Enterobacter* are motile and produce ornithine decarboxylase except *Enterobacter agglomerans* (4). Decarboxylase media are also recommended by standard methods for identification of bacteria (5-8).

This medium contains beef extract and peptic digest of animal tissue which provide nitrogenous nutrients for the growth of bacteria. Dextrose is the fermentable carbohydrate and pyridoxal is the co-factor for the decarboxylase enzyme. Bromo cresol purple and cresol red are the pH indicators in this medium. When the medium is inoculated with the dextrose fermenting bacteria, the pH is lowered due to acid production which changes the colour of the indicator from purple to yellow.

Acid produced stimulates decarboxylase enzyme. Putrescine is produced due to ornithine decarboxylation. Formation of amine putrescine increases the pH of the medium, changing the colour of the indicator from yellow to purple. If the organisms do not produce the appropriate enzyme, the medium remains acidic, yellow in colour. Each isolate to be tested should also be inoculated into the basal medium tube lacking the amino acid. After incubation, a decarboxylase test may show two layers of different colours, yellow and purple. Shake the tube gently before interpreting the results (4).

Inoculated tubes must be protected from air with a layer of sterile mineral oil. Exposure to air may cause alkalinization at the surface of the medium which makes the test invalid.

Quality Control

Appearance

Light yellow to greenish yellow homogeneous free flowing powder

Colour and Clarity of prepared medium

Purple coloured clear solution without any precipitate

Reaction

Reaction of 2.05% w/v aqueous solution at 25°C. pH : 6.0±0.2

pH

5.80-6.20

Cultural Response

M688: Cultural characteristics observed after an incubation at 35-37°C for upto 4 days (Inoculated tubes are overlaid with sterile mineral oil).

Organism	Inoculum (CFU)	Ornithine decarboxylation
<i>Citrobacter freundii</i> ATCC 8090	50-100	variable reaction
<i>Enterobacter aerogenes</i> ATCC 13048	50-100	positive reaction, purple colour
<i>Escherichia coli</i> ATCC 25922	50-100	variable reaction
<i>Klebsiella pneumoniae</i> ATCC 13883	50-100	negative reaction, yellow colour
<i>Proteus mirabilis</i> ATCC 25933	50-100	positive reaction, purple colour
<i>Proteus vulgaris</i> ATCC 13315	50-100	negative reaction, yellow colour
<i>Pseudomonas aeruginosa</i> ATCC 9027	50-100	negative reaction, yellow colour
<i>Salmonella Paratyphi A</i> ATCC 9150	50-100	positive reaction, purple colour
<i>Salmonella Typhi</i> ATCC 6539	50-100	negative reaction, yellow colour
<i>Serratia marcescens</i> ATCC 8100	50-100	positive reaction, purple colour
<i>Shigella dysenteriae</i> ATCC 13313	50-100	negative reaction, yellow colour
<i>Shigella flexneri</i> ATCC 12022	50-100	negative reaction, yellow colour
<i>Shigella sonnei</i> ATCC 25931	50-100	positive reaction, purple colour

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

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4. MacFaddin J., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd ed., Williams and Wilkins, Baltimore.
5. Isenberg (Ed.), 1992, Clinical Microbiology Procedures Handbook, Vol. I, ASM, Washington, D. C.
6. FDA Bacteriological Analytical Manual, 8th Ed., AOAC International, Gaithersburg, Md.
7. Eaton A. D., Clesceri L. S. and Greenberg A. E., (Ed.), 1998, Standard Methods for the Examination of Water and Wastewater, 20th Ed., American Public Health Association, Washington, D.C

8. Downes F. P. and Ito K., (Ed.), 2001, Compendium of Methods for the Microbiological Examination of Foods, 4th Ed., American Public Health Association, Washington, D.C

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