



*Medium 18.Urea Broth Medium

MM111

Urea Broth Medium is recommended for the identification of bacteria on the basis of urea utilization, specifically for the differentiation of *Proteus* species from *Salmonella* and *Shigella* species in accordance with Indian Pharmacopoeia, 2007.

Composition**

Ingredients	Gms / Litre
Potassium dihydrogen orthophosphate	9.100
Yeast extract	0.100
Anhydrous disodium hydrogen phosphate	9.500
Urea	20.000
Phenol red	0.010

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 38.71 grams in 1000 ml purified/ distilled water. Mix thoroughly to dissolve the medium completely. Sterilize by filtration. Aseptically dispense in sterile tubes as desired.

Principle And Interpretation

Urea Broth Medium was developed by Rustigian and Stuart (1). This medium is especially recommended by Indian Pharmacopoeia (5) for the differentiation of *Proteus* species from *Salmonella* and *Shigella* species in the enteric infection diagnosis (2), based on urea utilization (3, 4). It is also recommended for microbial limit tests. Other Gram-negative enteric bacilli are unable to utilize urea and fails to grow because of reduced availability of other nutrients. .

Urea Broth Medium becomes alkaline as the utilization of urea by the organisms liberate ammonia during the incubation, indicated by pink red colour. All urea test media rely on the alkalinity formation and so they are not specific for urease testing. Yeast extract provides essential vitamins and other growth factors. Phosphates aids as good buffering agent.

The utilization of proteins may raise the pH to alkalinity due to protein hydrolysis and excess of amino acids results in false-positive reaction. This medium shows positive reaction with Genus *Proteus* , few *Providencia* and *Morganella* sp. species

Quality Control

Appearance

Light yellow to light pink homogeneous free flowing powder

Colour and Clarity of prepared medium

Yellow orange coloured clear solution

Growth Promotion Test

Growth Promotion is carried out in accordance with Indian Pharmacopoeia

Cultural Response

Cultural characteristics observed after an incubation at 36-38°C for 18-24 hours.

Cultural Response

Organism	Inoculum (CFU)	Urease
Cultural Response <i>Enterobacter aerogenes</i> ATCC 13048	50-100	Negative reaction, no change
<i>Salmonella Typhimurium</i> ATCC 14028	50-100	Negative reaction, no change

<i>Proteus vulgaris</i> ATCC 13315	50-100	Positive reaction, cerise colour
<i>Klebsiella pneumoniae</i> ATCC 13883	50-100	Positive reaction, cerise colour
<i>Escherichia coli</i> ATCC 8739	50-100	Negative reaction, no change
<i>Klebsiella pneumoniae</i> ATCC 10031	50-100	Positive reaction, cerise colour
<i>Escherichia coli</i> NCTC 9002	50-100	Negative reaction, no change

Storage and Shelf Life

Store below 8°C in tightly closed container and the prepared medium at 2-8°C. Use before expiry date on the label.

Reference

1. Rustigian and Stuart, 1941, Proc. Soc. Exp. Biol. Med., 47:108.
2. Forbes, B.A.; Sahm, D.F. and Weissfeld, A.S., 2002, Bailey and Scott's Diagnostic Microbiology, 11th ed., The C.V. Mosby Co., St. Louis.
3. Christensen, 1946, J. Bact., 52:461.
4. MacFaddin J., 2000, Biochemical Tests for Identification of Medical Bacteria, 3rd ed., Williams and Wilkins, Baltimore.
5. Indian Pharmacopoeia, 2007 Govt. of India, Ministry of Health and Family Welfare, New Delhi, India.

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