Rapid Urease Test Broth

M1828

Intended Use

Recommended for the differentiation of organisms, especially the *Enterobacteriaceae* on the basis of urease production.

Composition**

Ingredients	Gms / Litre
Yeast extract	0.100
Urea	20.000
Potassium dihydrogen phosphate	0.091
Disodium hydrogen phosphate	0.095
Phenol red	0.010
Final pH (at 25°C)	6.8±0.2

**Formula adjusted, standardized to suit performance parameters

Directions

Suspend 20.30 grams in 1000 ml purified / distilled water. Mix well and sterilize by filtration. DO NOT AUTOCLAVE OR HEAT THE MEDIUM. Dispense in sterile tubes or flasks as desired.

Principle And Interpretation

In Rapid Urease test Broth the urease reaction given by *H. pylori*, occurs more quickly than that seen by other organisms which may split urea. As a result, it is an effective presumptive test for the presence of *H. pylori*. It is also used for the rapid detection of urease activity in bacteria such as *Proteus* spp., or in yeast, (such as *Cryptococcus neoformans*).

Helicobacter pylori is a gram negative, curved, microaerophilic and motile organism with multiple polar flagella.

Helicobacter pylori is a spiral urease producing organism that lies in the interface between gastric epithelial cell surface and the overlying mucus gel (5). It resides in the stomach of man and other primates, lining up the gastric mucus secreting cells. Rapid urease test is one of the invasive tests. This method has been used to help simplify the diagnosis of *H. pylori*, especially those specimens originating from duodenal and gastric ulcers, and chronic antral gastritis (type B).

This medium is develop as per McFaddin (4). Urease activity can be described as the splitting of urea via hydrolysis by a urease enzyme. The end products from this reaction yield ammonium carbonate and ammonia, which are alkaline in nature. The consequent rise in the pH of the medium is detected by phenol red indicator. The test is non-toxic, and the pH change that occurs from accumulation of alkaline end products is detected by a pH indicator in the media (3). *Helicobacter pylori* is an organism that may be easily identified by this test because of its very high endogenous urease activity.

Yeast extract which provides nitrogen and vitamin required for growth. Phosphates serve to buffer the medium.

Type of specimen

Isolated Microorganism

Specimen Collection and Handling

For isolated Microorganism, follow appropriate techniques for sample collection, processing as per guidelines and local standards(4). 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.The Urease test is pH dependant, accumulation of alkaline end products may interfere with the positive results
2. Further biochemical and serological testing is required 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

Light yellow to light pink homogeneous free flowing powder

Colour and Clarity of prepared medium

Yellowish orange coloured clear solution in tubes.

Reaction

Reaction of basal medium (1.87gm in 100ml distilled water) at 25°C. pH : 6.8±0.2

pH

6.60-7.00

Cultural Response

Cultural characteristics observed after an incubation at 35-37°C for 4-18 hours.

Organism	Inoculum (CFU)	Urease
# *Klebsiella aerogenes ATCC 13048* (00175*)	50-100	negative reaction, no change
Escherichia coli ATCC 25922 (00013*)	50-100	negative reaction, no change
Klebsiella pneumoniae ATCC 13883 (00097*)	50-100	weak positive reaction
Proteus vulgaris ATCC 13315	50-100	positive reaction, cerise colour
Salmonella Typhimurium *ATCC 14028* (00031*)	50-100	negative reaction, no change
Helicobacter pylori ATCC 43504	50-100	positive reaction, cerise colour
Klebsiella pneumoniae ATCC 10031	50-100	weak positive reaction

Key : (*) Corresponding WDCM numbers. (#) Formerly known as *Enterobacter aerogenes*

Storage and Shelf Life

Store dehydrated 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 clinical sample must be decontaminated and disposed of in accordance with current laboratory techniques (1,2).

Reference

1. Isenberg, H.D. Clinical Microbiology Procedures Handbook. 2nd Edition.
2. 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.
3. Klein PD, Graham DY, Gaillour A et al water source as risk factor for *Helicobacter pylori* infection in Peruvian children.Lancet 1991 ; 337 : 1503-06.

4. MacFaddin, Jean F-Biochemical tests for identification of medical bacteria / Jean F. Macfaddin1980; 424.

5. Mendall MA, Pajares-Garcia Epidemiology and transmissin of *Helicobacter pylori* . Curr Opin Gasteroenterol 1995; 11(supp 1) : 1-4.

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