UREA BROTH (STUART'S)

INTENDED USE

Remel Urea Broth (Stuart's) is a liquid medium recommended for use in qualitative procedures to aid in differentiation of genera within the *Enterobacteriaceae* on the basis of urease production.

SUMMARY AND EXPLANATION

Urea Broth (Stuart's) was developed by Stuart et al. for differentiation of *Proteus* from other genera within in the *Enterobacteriaceae*.^{1,2} This medium is designed for detection of urease activity in organisms which produce ammonia in a highly buffered medium with limited nutrients. Such organisms include all *Proteus* spp., *Morganella morganii*, *Providencia rettgeri*, and urease-positive *Providencia stuartii*.³ Other genera within the *Enterobacteriaceae* which hydrolyze urea, will not do so in this medium.

PRINCIPLE

Urea Broth (Stuart's) is a medium with a high buffering capacity and limited nutrients. Yeast extract provides essential growth factors required by strong urease-positive organisms, such as *Proteus* spp., *M. morganii*, and some species of *Providencia*. Organisms which possess the urease enzyme hydrolyze urea and produce an alkaline by-product (ammonia) which is detected by a change in the phenol red indicator from yellow-orange to pink.

REAGENTS (CLASSICAL FORMULA)*

Urea	g	Yeast Extract	0.1	g
Disodium Phosphate	g	Phenol Red	0.01	g
Monopotassium Phosphate9.1	g	Demineralized Water 1	000.0 n	nl

pH 6.8 ± 0.2 @ 25°C

*Adjusted as required to meet performance standards.

PROCEDURE

- 1. Inoculate Urea Broth (Stuart's) heavily (3 loopfuls) from an 18-24 hour, pure culture of the test isolate.
- 2. Shake the tube gently to suspend the bacteria.
- 3. Incubate aerobically with cap loosened at 33-37°C in an incubator or water bath.
- 4. Observe for a pink to red color development after 8, 12, 24, and 48 hours.

INTERPRETATION OF THE TEST

Positive Test - An intense pink-red color throughout the broth Negative Test - No color change; broth remains yellow-orange

QUALITY CONTROL

All lot numbers of Urea Broth (Stuart's) have been tested using the following quality control organisms and have been found to be acceptable. Testing of control organisms should be performed in accordance with established laboratory quality control procedures. If aberrant quality control results are noted, patient results should not be reported.

CONTROL	
Proteus mirabilis ATCC® 12453	3
Escherichia coli ATCC [®] 25922	

INCUBATION Aerobic, 18-24 h @ 33-37°C Aerobic, 18-24 h @ 33-37°C RESULTS Positive Negative

LIMITATIONS

- 1. Urea test media rely on demonstration of alkalinity and are not specific for detection of urease activity. Peptones in the medium may be hydrolyzed releasing amino acid residues, raising the pH, and resulting in false-positive reactions. A control test using the same test medium without urea can be used to facilitate interpretation of questionable reactions.³
- 2. The high buffering system in this medium masks urease activity in organisms that are delayed positive. Therefore, it is designed only for *Proteus* species, *Morganella morganii*, *Providencia rettgeri*, and urea-positive *Providencia stuartii*.³
- 3. False-negative results may occur if the amount of ammonia formed is too slight to change the hydrogen ion concentration of the phenol red indicator to an alkaline pH; in such instances, the broth will remain yellow-orange in color.⁴

BIBLIOGRAPHY

- 1. Rustigian, R. and C.A. Stuart. 1941. Proc. Soc. Exp. Biol. Med. 47:108-112.
- 2. Stuart, C.A., E. Van Stratum, and R. Rustigian. 1945. J. Bacteriol. 49:437-444.
- 3. MacFaddin, J.F. 1985. Media for Isolation-Cultivation-Identification-Maintenance of Medical Bacteria. Vol. 1. Williams & Wilkins, Baltimore, MD.
- 4. MacFaddin, J.F. 2000. Biochemical Tests for Identification of Medical Bacteria. 3rd ed. Lippincott Williams & Wilkins, Philadelphia, PA.

Refer to the front of Remel *Technical Manual of Microbiological Media* for **General Information** regarding precautions, product storage and deterioration, specimen collection, storage and transportation, materials required, quality control, and limitations.

 ATCC^{\otimes} is a registered trademark of American Type Culture Collection. IFU 65230, Revised September 2, 2009

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