Urease - Indole

REF CK4651

Intended Use

The urease and indole tests are commonly used for the identification of Enterobacterales and non-fermenting Gram negative bacteria

Background

The urease – Indole test is used to screen for potential Salmonella and Shigella colonies on enteric culture media.

The urease test will determine the ability of an organism to split urea, through the production of the enzyme urease. Two units of ammonia are formed with resulting alkalinity in the presence of the enzyme, and the increased pH is detected by a pH indicator.

The Indole test identifies organisms capable of degrading the amino acid tryptophan which results in the release of indole. The addition of Kovacs Reagent produces a deep red colour ring on the surface of the tube in the presence of indole.

Salmonellae and Shigellae are urease negative whilst Enterobacterales such as *Proteus, Morganella, Klebsiella* and some *Citrobacter* are strongly urease positive. The production of indole from tryptophan is a characteristic absent in *Salmonella* but present in *Escherichia coli, Morganella* and some species of *Klebsiella, Aerobacter* and *Citrobacter*.

The test involves preparing a heavy suspension of the test organism in the presence of urea and tryptophan. Following incubation the development of a pink or reddish colour is a positive reaction for urease, the development of a red surface ring following the addition of Kovacs Reagent is a positive reaction for indole.

Precautions

This product is for in-vitro diagnostic use and should be used by properly trained laboratory professionals. Universal precautions should be taken in the handling, processing and discarding of all materials used to perform the test. Do not use reagents after the expiration date shown on the product label has expired.

Methods

Use a fresh 18-24 hour culture as older cultures could be less metabolically active and results from these may be unreliable.

Tuhe:

Add 3 to 5 drops of sterile distilled water to the urease - indole test tube.

Using a loop, wire or stick take a heavy inoculum (McFarland No. 4) of the test organism and thoroughly mix in the urease – indole test tube.

Incubate at 37°C for 4 hours or 18-24 hours, following incubation observe the colour of the suspension, a pink or reddish colour is a positive result for urease.

To perform the indole test (once the urease test result has been recorded) add 2 drops of

Kovacs Reagent to the tube. Leave to stand for 3 to 4 minutes before shaking. A positive test will develop a deep red surface layer (look only at the colour of the surface layer).

Results

Urease

Positive Reaction – pink or reddish colour Negative Reaction – straw, peach, yellow or colourless.

Indole

Positive Reaction – surface layer will be a red colour

Negative Reaction – surface layer will be a yellow or straw colour

Limitations

This test should be performed on microorganisms grown on media appropriate to the specimen.

Quality Control

A quality control should be undertaken daily or immediately prior to use.

Positive control (URE pos., IND pos.)-Morganella morganii ATCC 25830 Positive control (URE pos., IND neg.)-Klebsiella pneumoniae ATCC 13883

Negative control (URE neg., IND pos.)-Escherichia coli ATCC 25922

Shelf Life & Storage

The expiry date, storage temperature (ambient) and storage conditions are indicated on the outer package label.

Materials provided

Each pack contains 28 capped tubes, each tube includes a tablet containing 5mg of urea and 2.5mg of tryptophan.

Materials required but not provided

Sterile loops or needles Sterile distilled water Kovacs Reagent (CK9020)

References

Standards Unit, National Infection Service, PHE. UK SMI, TP36-Urease Test, Issue 4, 02.04.19.

Standards Unit, National Infection Service, PHE. UK SMI, TP19-Indole Test, Issue 4, 03.12.18.

Barrow, G.I. & Feltham, R.K.A. Cowan and Steel's Manual for the Identification of Medical Bacteria. Third edition.

Ford, M. Medical Microbiology. Oxford University Press.

REF	Catalogue number	
LOT	Batch number	
Σ	Use by date	
IVD	In-Vitro Diagnostic device	
Σ	Contains sufficient for <n> tests</n>	
1	Temperature storage limitations	
[]i	Consult instructions for use	
***	Manufacturer	

Issue	Date	Comments
4	08/09/2020	IFU format revision.

