remel

Spot Indole Reagent

INTENDED USE
Remel Spot Indole Reagent is recommended for use in qualitative procedures to determine the ability of an organism to split indole from the tryptophan molecule.

SUMMARY AND EXPLANATION
Vracko and Sherris, in 1963, utilized Spot Indole Reagent for the presumptive separation of the *Proteus* species and *Escherichia coli*.[1] In 1969, Lowrance, Reich, and Traub found ρ-Dimethyl-aminocinnamaldehyde to be the most sensitive indole reagent, capable of detecting 3 mcg of indole per milliliter of medium.[2]

PRINCIPLE
Intracellular enzymes collectively called "typtophanases" mediate the production of indole by hydrolytic activity against the amino acid tryptophan. Indole combines with dimethyl-aminocinnamaldehyde to form a blue-green compound. The reaction occurs by a condensation process formed by an acid splitting of the protein.

REAGENTS (CLASSICAL FORMULA)*

ρ-Dimethylaminocinnamaldehyde (CAS 6203-18-5)	10.0 g
Hydrochloric Acid (Conc.) (CAS 7647-01-0)	100.0 ml
Demineralized Water (CAS 7732-18-5)	900.0 ml

*Adjusted as required to meet performance standards.

PRECAUTIONS
DANGER! POISON, may be harmful or fatal if swallowed**. CORROSIVE,** may cause burns or irritation to skin, eyes, or respiratory tract.

This product is for *In Vitro Diagnostic Use* and should be used by properly trained individuals. Precautions should be taken against the dangers of microbiological hazards by properly sterilizing specimens, containers, and media after their use. Directions should be read and followed carefully. Refer to Material Safety Data Sheet for additional information.

STORAGE
This product is ready for use and no further preparation is necessary. Store product in its original container at 2-30°C until used. Allow product to come to room temperature before use. Do not incubate prior to use. Protect product from light.

PRODUCT DETERIORATION
This product should not be used if (1) the color has changed, (2) the expiration date has passed, or (3) there are other signs of deterioration.

SPECIMEN COLLECTION, STORAGE AND TRANSPORT
Specimens should be collected and handled following recommended guidelines.[3]

MATERIALS REQUIRED BUT NOT SUPPLIED
(1) Loop sterilization device, (2) Inoculating loop, swab, collection containers, (3) Incubators, alternative environmental systems, (4) Supplemental media, (5) Quality control organisms, (6) Whatman (No. 1) filter paper.

PROCEDURE
Filter Paper Method:
1. Dispense 1 or 2 drops of reagent onto a piece of Whatman (No. 1) filter paper or equivalent.
2. Smear the growth from an actively growing pure culture onto the saturated filter paper.
3. Observe for the development of a blue color within 1 to 3 minutes**.**

Swab Method:
1. Dispense 1 or 2 drops of reagent onto the tip of a cotton swab.
2. Touch the tip of the saturated swab to the top of a test colony growing on a culture medium.
3. Observe for the development of a blue color within 1 to 3 minutes.

INTERPRETATION
Positive Test - Blue color development within 3 minutes
Negative Test - Pink color development

QUALITY CONTROL
All lot numbers of Spot Indole Reagent have been tested using the following quality control organisms and have been found to be acceptable. Testing of a positive and negative control 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	RESULTS
Bacteroides ovatus ATCC® 8483	Positive
Escherichia coli ATCC® 25922	Positive
Prevotella melaninogenica ATCC® 25845	Negative
Proteus mirabilis ATCC® 12453	Negative

LIMITATIONS

1. Test only colonies cultured on non-glucose containing media, as glucose inhibits indole production.
2. Organisms from MacConkey Agar and EMB Agar cannot be tested; indicators in these media may cause a false positive reaction.
3. Certain strains of *Proteus vulgaris*, *Providencia* spp., and *Aeromonas* spp. will give a false negative reaction with the Spot Indole test.[4]
4. Media utilized in this test should be checked with known positive and negative control organisms to ensure adequate tryptophan content necessary for the indole reaction.
5. Because adjacent colonies are likely to take up diffused indole, positive tests are valid only if pure cultures are tested.[5]

BIBLIOGRAPHY

1. Vracko, R. and J.C. Sherris. 1963. Am. J. Clin. Path. 39:429-432.
2. Lowrance, B.L., P. Reich, and W.H. Traub. 1969. Appl Microbiol. 17:923-924.
3. Murray, P.R., E.J. Baron, J.H. Jorgensen, M.A. Pfaller, and R.H. Yolken. 2003. Manual of Clinical Microbiology, 8th ed. ASM, Washington, D.C.
4. Balzevic, D.J. and G.M. Ederer. 1975. Principles of Biochemical Tests in Diagnostic Microbiology. John Wiley & Sons, New York, NY.
5. Sutter, V.L. and W.T. Carter. 1972. Am. J. Clin. Path. 58:335-338.

PACKAGING

REF 21245..... 25 ml/Btl

Symbol Legend

REF	Catalog Number
IVD	In Vitro Diagnostic Medical Device
LAB	For Laboratory Use
	Consult Instructions for Use (IFU)
	Temperature Limitation (Storage Temp.)
LOT	Batch Code (Lot Number)
	Use By (Expiration Date)

ATCC® is a registered trademark of American Type Culture Collection.
CAS (Chemical Abstracts Service Registry No.)

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