remel

TB BRILLIANT GREEN

INTENDED USE
Remel TB Brilliant Green is a reagent recommended for use in qualitative procedures as a counterstain to differentiate acid-fast bacteria from nonacid-fast bacteria.

SUMMARY AND EXPLANATION
The microscopic acid-fast staining technique is one of the earliest methods used for detection of the tubercle bacillus.[1-3] Because the acid-fast stain remains the most rapid method for detection of mycobacteria it continues to be an invaluable adjunct to culture in clinical microbiology laboratories.[4] The carbolfuchsin staining technique is also used to detect partially acid-fast organisms such as *Rhodococcus* and *Nocardia*.

PRINCIPLE
Mycolic acids and waxes in the cell wall of the acid-fast organism complex with carbolfuchsin, a basic dye, which is retained in the cell wall after mild acid decolorization. Typical acid-fast organisms stain purple to red. TB Brilliant Green is used as the counterstain to detect nonacid-fast bacteria, which stain green.

REAGENTS (CLASSICAL FORMULA)*
Brilliant Green (CAS 633-03-5).....5.0 g
Demineralized Water (CAS 7732-18-5).....1000.0 ml

*Adjusted as required to meet performance standards.

PRECAUTIONS
Caution! May cause eye, skin, and respiratory tract irritation. This product is considered a low hazard for usual industrial handling.

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, test materials, and media after use. Directions should be read and followed carefully. Refer to Material Safety Data Sheet for additional information on reagent chemicals.

STORAGE
This product is ready for use and no further preparation is necessary. Store product in its original container at room temperature (20-25°C) until used.

PRODUCT DETERIORATION
This product should not be used if (1) the color has changed from dark green, (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.[4-6]

MATERIALS REQUIRED BUT NOT SUPPLIED
(1) Loop sterilization device, (2) Inoculating loop, swabs, collection containers, (3) Incubators, alternative environmental systems, (4) Supplemental media, (5) TB Kinyoun Carbolfuchsin (REF R40104), (6) TB Decolorizer (REF R40106), (7) Bunsen burner or slide warmer, (8) Microscope, glass slides, immersion oil, (9) Quality control organisms.

PROCEDURE
Every specimen represents a potential source of infectious material and should be handled accordingly.[4-6]

1. Make a thin smear of the material for study and allow it to air dry. Heat fix by passing the slide through the flame of a Bunsen burner or use a slide warmer.
2. Flood the smear with TB Kinyoun Carbolfuchsin and stain for 5 minutes. Rinse with water and drain.
3. Decolorize with TB Decolorizer for 3 minutes. Rinse with water and drain.
4. Repeat decolorization for 1-2 minutes or until no red stain flows from the smear. Rinse with water and drain.
5. Flood smear with TB Brilliant Green and stain for 3-4 minutes. Rinse with water and allow to air dry.
6. Examine under oil immersion objective (100 X) for purple to red acid-fast bacilli.

INTERPRETATION
Positive Test - Mycobacteria stain purple to red and are small, slightly curved rods, possibly beaded or banded, with tapered ends.
Negative Test - Nonacid-fast organisms stain green.

QUALITY CONTROL
All lot numbers of TB Brilliant Green have been tested and found to be acceptable. Testing of control organisms should be performed in accordance with the quality control procedures established by each laboratory following applicable regulatory agencies. If aberrant quality control results are noted, patient results should not be reported.

Acid-Fast Stain for Mycobacteria: Positive and negative control slides should be included every time the acid-fast stain is performed for detection of mycobacteria.[4]

LIMITATIONS
1. Examine a minimum of 300 oil immersion fields before reporting as negative.[4]
2. Nontuberculous *Mycobacterium* strains (e.g., *M. avium* complex) retain the basic dye and appear acid-fast; however, such strains are usually morphologically atypical (i.e., pleomorphic or coccoid). Positive acid-fast smear reports should be based only on typical forms, but atypical cells should be noted.[5,6]
3. Atypical rods may represent partially acid-fast organisms, such as *Nocardia* or *Rhodococcus.* A weaker acid or a shorter destaining period should be used to detect these organisms.[5]
4. The sensitivity of the direct acid-fast smear examination for the diagnosis of mycobacterial infection is lower than that of culture methods. Cultures should be performed on all specimens.[4,6]

BIBLIOGRAPHY

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3. Bishop, P.J. and G. Neumann. 1970. Tubercle. 51:196-206.
4. Isenberg, H.D. 2004. Clinical Microbiology Procedures Handbook. 2nd ed. ASM Press, Washington, D.C.
5. Murray, P.R., E.J. Baron, J.H. Jorgensen, M.L. Landry, and M.A. Pfaller. 2007. Manual of Clinical Microbiology. 9th ed. ASM Press, Washington, D.C.
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PACKAGING
REF R40100, TB Brilliant Green.....250 ml/Btl
REF R40200, TB Brilliant Green..... 250 ml/Btl, 5/Pk

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)

CAS (Chemical Abstracts Service Registry No.)
Manufactured for Remel Inc.

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