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Fite's Stain Kit (For Leprosy & Nocardia)

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Intended Use
For In Vitro Diagnostic Use

Summary and Explanation
The Fite's Stain Kit (For Leprosy and Nocardia) is intended for use in the histological visualization of mycobacterium Lepra bacillus and Nocardia. This kit may be used on formalin-fixed, paraffin-embedded or frozen sections.

Lepra bacillus: Red
Nocardia: Red
Background: Blue

Control Tissue
Tissues fixed in 10% formalin are suitable for use prior to paraffin embedding. Consult references (Kiernan, 1981: Sheehan & Hrapchak, 1980) for further details on specimen preparation.

1. Cut sections, usually 3 to 5 µm and pick the sections up on glass slides.
2. Bake the slides for at least 30 minutes at approximately 70°C.
3. Allow to cool.

Reagents Provided

Kit Contents	Volume	Storage
Xylene-Peanut Oil Solution	125 mL	15-30°C
Carbol Fuchsin Solution	125 mL	15-30°C
Acid Alcohol Solution (1%)	500 mL	15-30°C
Methylene Blue Solution	125 mL	15-30°C

Storage and Handling
Do not use product after the expiration date printed on vial. If reagents are stored under conditions other than those specified here, they must be verified by the user. Diluted reagents should be used promptly.

Lepra bacillus Procedure

1. Deparaffinize sections in 2 changes of Xylene-Peanut Oil Solution for 12 minutes each.
2. Air dry slide for 15 minutes. Do not remove oil film. Remaining film prevents de-staining of Lepra bacillus during differentiation.
3. Rinse slide in several changes of distilled water.
4. Incubate slide in Carbol Fuchsin Solution for 15 minutes.
5. Rinse slide in several changes of distilled water.
6. Differentiate section in Acid Alcohol Solution (1%) until background is pink.
7. Rinse slide in distilled water and check by microscope for correct differentiation.
8. Rinse in running tap water for 1 minute followed by 1 rinse in distilled water.
9. Dip slide 2-3 times in Methylene Blue Solution.
10. Dip slide quickly in distilled water and check by microscope for correct staining.
11. Air dry slide at room temperature.
12. Dip slide several times in Xylene or Xylene Substitute
13. Mount in synthetic resin.

Nocardia Procedure

Prepare the Following Solutions Immediately Before Use

1. Prepare Diluted Acid Alcohol Solution by mixing 25ml of Acid Alcohol Solution (1%) with 25ml of Distilled Water.

Staining Procedure

1. Deparaffinize sections in 2 changes of Xylene-Peanut Oil Solution for 12 minutes each.
2. Air dry slide for 15 minutes. Do not remove oil film. Remaining film prevents de-staining of Lepra bacillus during differentiation.
3. Rinse slide in several changes of distilled water.
4. Incubate slide in Carbol Fuchsin Solution for 15 minutes.
5. Rinse slide in several changes of distilled water.
6. Dip slide 2-3 times in Diluted Acid Alcohol Solution.
7. Rinse slide in distilled water and check by microscope for correct differentiation. Avoid decolorizing the Nocardia organism.
8. Rinse in running tap water for 1 minute followed by 1 rinse in distilled water.
9. Dip slide 2-3 times in Methylene Blue Solution.
10. Dip slide quickly in distilled water and check by microscope for correct staining.
11. Air dry slide at room temperature.
12. Dip slide several times in Xylene or Xylene Substitute.
13. Mount in synthetic resin.

Limitations of the Procedure

1. Histological staining is a multiple step diagnostic process that requires specialized training in the selection of the appropriate reagents, tissue selections, fixation, processing, preparation of the slide, and interpretation of the staining results.
2. Tissue staining is dependent on the handling and processing of the tissue prior to staining.
3. Improper fixation, freezing, thawing, washing, drying, heating, sectioning, or contamination with other tissues or fluids may produce artifacts or false negative results.
4. The clinical interpretation of any positive staining, or its absence, must be evaluated within the context of clinical history, morphology and other histopathological criteria. It is the responsibility of a qualified pathologist to be familiar with the special stain and methods used to produce the slide.
5. Staining must be performed in a certified licensed laboratory under the supervision of a pathologist who is responsible for reviewing the stained slides and assuring the adequacy of positive and negative controls.

Precautions

1. Consult local and/or state authorities with regard to recommended method of disposal.
2. Materials of human or animal origin should be handled as biohazardous materials and disposed of with proper precautions.
3. Avoid microbial contamination of reagents. Contamination could produce erroneous results.
4. This reagent may cause irritation. Avoid contact with eyes and mucous membranes.
5. If reagent contacts these areas, rinse with copious amounts of water.
6. Do not ingest or inhale any reagents.

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Troubleshooting

If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem is suspected, contact Diagnostic BioSystems Technical Support at (925) 484-3350, extension 2 or techsupport@dbiosys.com.

References

I. Fite, G.L. Cambre, P.J., and Turner, M.H. Procedure for demonstrating lepra bacilli I paraffin sections. Arch. Pathol., Vol. 43, pages 624-25, 1947.

II. Mallory, Pathological Technique; page 275.

III. Clarke, G., et al. Staining Procedures, 4th Edition, Williams& Wilkins, page 447, 1981.

IV. Crowder, C., Taylor, HW, Modified Fite Stain for Demonstration of Mycobacterium Species in Tissue Sections; Journal of Histotechnology; Volume 19; 2: pages 133-134.

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