

## AURAMINE O RHODAMINE B METHOD FOR AFB FLUORESCENT STAIN

<b>PURPOSE:</b>	For In Vitro Diagnostic Use: Intended for the qualitative demonstration of mucin or Cryptococcus.
<b>PRINCIPLE:</b>	The staining solution, along with the aid of heat, is able to dissolve into the lipid capsule of the bacteria and then resist being destained by the acid which removes all unbound stain. The dyes fluoresce under UV light.
<b>CONTROL:</b>	Any tissue known positive for acid fast organisms <i>Control Slides can be purchased from Histology Control Systems. See inside back cover, Item# cs003.</i>
<b>SPECIMEN PREPARATION:</b>	Formalin fixed, paraffin embedded sections cut at 6 micrometers
<b>SOLUTIONS:</b>	1. Auramine Rhodamine Item# s119B 2. Hydrochloric Acid 0.5% in 70% Alcohol Item# s2194 3. Potassium Permanganate 0.5% Aqueous Item# s263  <i>Solutions can be purchased separately from Poly Scientific.</i>
<b>NOTES:</b>	
<b>REFERENCE:</b>	Kuper, S.W.A., May, J.R. "Detection of Acid Fast Organisms in Tissue Sections By Fluorescence Microscopy". <u>J. Path. Bact.</u> 1960; 79: pp. 59-68.

### STAINING PROCEDURE:

1. Deparaffinize and hydrate to distilled water.
2. Place in preheated Auramine Rhodamine in the oven at 60°C for 10 minutes.
3. Wash in tap water for 2 minutes.
4. Decolorize for 1-2 minutes in Hydrochloric Acid 0.5% in 70% Alcohol. (For Hansen's bacilli, it is recommended that decolorization be done with Hydrochloric Acid 0.5% Aqueous).
5. Wash in water for 2 minutes.
6. Differentiate in Potassium Permanganate 0.5% Aqueous for 2 minutes.
7. Wash in water for 2 minutes.
8. Dehydrate, clear, and mount with Poly Mount (Item# s2153) or any other acceptable mounting medium.

### RESULTS:

Acid Fast Organisms ..... Should show fluorescence  
 Non Acid Fast Organisms ..... Little or no fluorescence

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