

**WILDER'S METHOD FOR RETICULUM**

<b>PURPOSE:</b>	For In Vitro Diagnostic Use: Intended for the qualitative demonstration of reticulum fibers.
<b>PRINCIPLE:</b>	Phosphomolybdic Acid oxidizes the tissue and Uranyl Nitrate sensitizes the reticulum. The Diamino Silver Solution is taken up selectively by reticulum. The Reducing Solution then precipitates the visible metallic silver, old Chloride tones and Thiosulfate removes excess silver.
<b>CONTROL:</b>	Normal liver  <i>Control Slides can be purchased from Histology Control Systems. See inside back cover, Item# cs024.</i>
<b>SPECIMEN PREPARATION:</b>	Formalin fixed, paraffin embedded sections cut at 6 micrometers
<b>SOLUTIONS:</b>	<ol style="list-style-type: none"> <li>1. Phosphomolybdic Acid 10% Item# s2176</li> <li>2. Uranyl Nitrate 1% Aqueous Item# s288B</li> <li>3. Ammoniacal Silver Nitrate Solution Item# s114</li> <li>4. Gold Chloride 0.2% Aqueous Item# s202</li> <li>5. Sodium Thiosulfate 5% Aqueous Item# s1895</li> <li>6. Nuclear Fast Red Kernechtrot 0.1% Item# s248</li> <li>7. Solutions for Reducing Solution:                  Uranyl Nitrate 1% Aqueous..... 3 mL                  Formaldehyde 37% ..... 1 mL                  Distilled Water ..... 100 mL</li> </ol> <p><i>Solutions can be purchased separately from Poly Scientific.</i></p>
<b>NOTES:</b>	
<b>REFERENCE:</b>	Wilder, H.C. <u>Amer. J. Path.</u> 11: 817-821, 1935.

**STAINING PROCEDURE:**

1. Deparaffinize and hydrate to distilled water.
2. Oxidize in Phosphomolybdic Acid 10% for 1 minute.
3. Rinse well in running water or cells will hold yellow.
4. Sensitize in Uranyl Nitrate 1% Aqueous for 1 minute.
5. Rinse in distilled water for 10-20 seconds.
6. Place in Ammoniacal Silver Nitrate Solution for 1 minute.
7. Dip very quickly in 95% Alcohol and go immediately into Reducing Solution for 1 minute.
8. Rinse well in distilled water.
9. Tone in Gold Chloride 0.2% Aqueous for 10-60 seconds or until sections turn red. Check individually under microscope.
10. Rinse in distilled water.
11. Place in Sodium Thiosulfate 5% Aqueous for 1 minute.
12. Wash well in tap water.
13. Counterstain if desired with Nuclear Fast Red Kernechtrot 0.1%. Rinse well with distilled water.
14. Dehydrate in 95% Alcohol, Absolute Alcohol and clear in Xylene, 2 changes each.
15. Mount with Poly Mount (Item# s2153) or any other acceptable mounting medium.

**RESULTS:**

Reticulum Fibers ..... Black  
 Collagen..... Rose  
 Other Tissue Elements ..... Red

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