Copper Stain
Histo24.01

Describe briefly the most recent revision made to this policy, procedure or protocol & why:
New Procedure

Purpose/Policy Statement:
For diagnostic purposes, to demonstrate copper.

Definitions:
• n/a

STEPS / KEY POINTS

PROCEDURE:
1. Deparaffinize slide in Xylene and hydrate through alcohols.
2. Rinse slide in Tap water.
3. **Place slide in Working Rhodanine Solution and heat in a microwave oven at full power for 25 seconds. Agitate container and microwave again at 20 seconds. Temperature should be about 73 °C. DO NOT BOIL. Carefully agitate slide, cap container and allow the solution to cool on benchtop to room temperature; agitate container periodically during cooling. After cooling, quickly examine slide using a microscope; repeat heating and cooling cycles as needed to reach desired copper stain intensity.
4. Rinse slide through 2 changes of Acetate Buffer for 1 minute each.
5. Place slide in Modified Mayer’s Hematoxylin for 5-10 seconds with agitation.
6. Rinse slide through 3 changes of Acetate Buffer Solution for 1 minute each.
7. Dehydrate slide through 3 changes of fresh Absolute Alcohol.
8. Clear slide through 3 changes of fresh Xylene.
9. Coverslip immediately using a permanent mounting media.

** OPTIONAL: Place slides in Working Rhodanine Solution for 18 hours at 37 °C; proceed to Step 4.

QUALITY CONTROL:
American MasterTech Scientific Recommended Control Slide: Human Liver with Copper or order control slides CSCOP25.

PROFICIENCY TESTING: College of American Pathologists HistoQIP.
RESULTS:
Copper: Light brown to red cytoplasmic granules
Nuclei: Blue

PROCEDURE NOTES:

SPECIMEN: Paraffin sections cut at 4 microns and mounted on charged glass microslides.

MATERIALS, REAGENTS: MICROWAVE COPPER STAIN KIT ITEM KTMCO

<table>
<thead>
<tr>
<th>KIT COMPONENTS INCLUDED:</th>
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<tbody>
<tr>
<td>25 ml. RHODANINE, STOCK</td>
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<tr>
<td>100 ml. MODIFIED MAYER’S HEMATOXYLIN</td>
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<tr>
<td>2 PINTS ACETATE BUFFER</td>
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**CAUTION: CORROSIVE, IRRITANT**
Wear appropriate protective equipment.

**SOLUTIONS**

**WORKING RHODANINE SOLUTION:**
Rhodanine Stock……………. 4ml
Acetate Buffer …………….. 46ml

Agitate Rhodanine Stock thoroughly and immediately pipette into Acetate Buffer. **FILTER**

**INSTRUMENTATION OR EQUIPMENT:** Automated H&E stainer, .Light Microscope

**Form Name & Number or Attachment Name (If Applicable):**
HISTO24.01

**Author Position:**
Lead Histologist

**Review/Revision Authority (Position Not Individual Name):**
Lead Histologist

**Expert Consultant Position/s (Not Individual Name/s):**
N/A

**References (Required for Clinical Documents):**
Lindquist RR: Studies on the Pathogenesis of Hepatolenticular II: Cytochemical methods for the location of copper; Arch Pathol; 87; 1969,370.

**Is there a Regulatory Requirement? Yes ☐ No ☐**
If yes, insert requirement information here:

**Review History (No Changes):**
N/A

**Revision History (Note changes in area under header):**
N/A

**Computer Search Words:**
N/A

**Policy, Procedure or Protocol Cross Reference Information:**