COPPER - RHODANINE METHOD

PURPOSE: To identify copper deposits, such as in Wilson's Disease, left in the liver.

PRINCIPLE: It has been suggested that the rhodanine demonstrates the protein to which the copper binds rather than the copper itself.

CONTROL: Fetal liver, or a known positive. Coverslip the tissue section with Permount, the other fast drying mounting medias leech out the staining reaction.

FIXATIVE: 10% formalin

TECHNIQUE: Cut paraffin sections 4-5 microns.

EQUIPMENT: Rinse all glassware in DI water, coplin jars, microwave oven, course filter paper.

REAGENTS:
Rhodamine
Stock Solution:
5-p-dimethylaminobenzylidene rhodanine 0.2 gm
Absolute alcohol 100.0 ml
Mix well. Refrigerate, solution is stable for 3 months.

CAUTION: Flammable. Avoid contact and inhalation.

Rhodamine Working Solution:
Stock Rhodamine 10.0 ml
Distilled water 40.0 ml
Shake stock solution before measuring, filter through course filter paper. Make solution fresh, discard after use.

CAUTION: Avoid contact and inhalation.

Hematoxylin:
Commercial Gill-3

SAFETY: Wear gloves, goggles and lab coat. Avoid contact and inhalation.
PROCEDURE:
1. Deparaffinize and hydrate to distilled water.
2. Working Rhodanine solution, microwave 70 power, 60 seconds. Allow slides to stand in solution for 30 - 60 minutes.
3. Wash in several changes of distilled water.
4. Hematoxylin, 1-3 minutes.
5. Wash in water.
7. Wash in water.
8. Dehydrate quickly through the alcohols, clear, and coverslip in PERMOUNT, or the coverslipper.

RESULTS:
Copper deposits: bright red to orange
Nuclei: blue

REFERENCES:
PROCEDURE CARD

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DATE:________________________

TECH:________________________

EXPIRATION:__________________