



VitroView™ Rhodanine Copper Stain Kit

SKU#: VB-3021

Description: It has been suggested that the rhodanine demonstrates the protein to which the copper binds rather than the copper itself. This kit is designed to identify copper deposits, such as in Wilson's disease, left in the liver.

Kit Contents:

VB-3021-1	Rhodanine Stock Solution	30 ml
VB-3021-2	Acetate Buffer Solution (pH 8.0)	500 ml×2
VB-3021-3	Mayer's Hematoxylin Solution	250 ml

Storage

Store Rhodanine Stock Solution at 2-8°C and store the others at room temperature.

Protocol

Things to do before staining: Prepare Working Rhodanine Solution

- Shake Rhodanine Stock Solution
- Mix Rhodanine Stock Solution 4ml with Acetate Buffer Solution (pH 8.0) 46 ml

Procedure:

1. Deparaffinize in xylene I for 6 minutes and II for 6 minutes.
2. Rehydrate
 - a. Ethanol 100% (2 minutes)
 - b. Ethanol 100% (2 minutes)
 - c. Ethanol 95% (2 minutes)
 - d. Ethanol 95% (2 minutes)
 - e. Ethanol 70% (2 minutes)
3. Rinse in DI water (2 minutes).
4. Stain in Working Rhodanine Solution (three options, please only choose one)
 - at 60°C for 1-2 hours, or
 - at 37°C for 18 hours, or
 - Microwave Modification: Place slides in a plastic Coplin jar containing Working Rhodanine Solution and microwave for 6 minutes at 70°C.
5. At the end of incubation (for both oven and microwave), to avoid unwanted slide precipitate, pour off warm Working Rhodanine Solution into a second Coplin jar, reserve and set aside.
6. Rinse slides well in 3 changes of Acetate Buffer Solution (pH8.0) for 1 minute each..
7. Check positive control slide microscopically to determine adequate copper/reddish brown development. *Return slides to reserved Working Rhodanine Solution if additional incubation is required (repeat step 4, 5 and 6).*
8. Stain in Mayer Hematoxylin Solution for 5 minutes

9. Rinse in tap water, 2×2min
10. Dehydrate with 2 changes of 95% Ethanol and 2 changes of 100% Ethanol (2 minute per change).
11. Clear with 3 changes of xylene (5 minute per change)
12. Mount coverslip onto glass slide with Permount or some other suitable organic mounting medium.

Control Tissue: Fetal liver or a known positive.

RESULTS:

Copper	Copper/reddish brown
Nuclei	Light blue

Note: This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Precautions: Handle with care. Avoid contact with eyes, skin and clothing. Do not ingest. Wear gloves.