



PERIODIC ACID-METHENAMINE SILVER STAIN KIT PROCEDURE

Item# **CUKTPAM**

(Revised 03/7/18)

**KIT
COMPONENTS
INCLUDED:**

100 ml 0.5% PERIODIC ACID
30 ml 7% SILVER NITRATE
100 ml 3% SODIUM THIOSULFATE

100 ml METHENAMINE SOLUTION
100 ml 0.2% GOLD CHLORIDE
100 ml LIGHT GREEN COUNTERSTAIN

PRINCIPLE: This kit demonstrates Reticulum Fibers and Basement Membrane.

SPECIMEN: Any well fixed paraffin embedded tissue cut at 5 to 6 microns.

QUALITY CONTROL: American MasterTech Scientific Recommended Control Slide:
Reticulum Fibers, CSR0125P.

PROCEDURE:

1. Deparaffinize slide using Xylene or a Xylene Substitute and hydrate through alcohols.
2. Rinse slide thoroughly in Tap water.
3. Rinse slide in 2 changes of Distilled water.
4. Place slide in **0.5% PERIODIC ACID** for 15 minutes.
5. Rinse slide in Distilled water for 1 minute.
6. Place slide in heated **Methenamine/Silver Solution** until golden-brown color develops (check every 20 to 30 minutes).
7. Rinse slide through 2 changes of heated Distilled water. Check under microscope. Repeat **Step 6** if needed.
8. Place slide in **0.2% GOLD CHLORIDE** for 1 minute.
9. Rinse slide thoroughly in Distilled water.
10. Place slide in **3% SODIUM THIOSULFATE** for 2 minutes.
11. Rinse slide in running Tap water for 3 minutes.
12. Place slide in **LIGHT GREEN COUNTERSTAIN** for 30 to 60 seconds.
13. Dehydrate slide through 3 changes of fresh Absolute Alcohol.
14. Clear slide through 3 changes of Xylene or Xylene Substitute.
15. Coverslip using a permanent mounting media.

SPECIAL INSTRUCTIONS (Methenamine/Silver Hyperthermal Formulation)

Refrigerate (**DO NOT FREEZE!**) the separate bottles of **METHENAMINE SOLUTION** and **7% SILVER NITRATE** as soon as possible. Store both reagents in Refrigerator.

Just before use, combine 1 ml. **7% SILVER NITRATE** for every 20 ml. of **METHENAMINE SOLUTION** used and mix well. Heat working solution to 50°C to 60°C using a waterbath or oven. Heat two changes of Distilled water to the same temperature.

RESULTS:

Reticulum fibers, basement membranes: **GRAY TO BLACK**
Background: **GREEN**

REFERENCE: Sheehan DC Hrapchak BB: Theory and Practice of Histotechnology; 1980, 187.

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