



## JONES STAIN KIT FOR BASEMENT MEMBRANE & RETICULUM

Item# **CUKTJON**

(Revised 03/7/18)

**KIT  
COMPONENTS  
INCLUDED:**

1 PINT 0.5% PERIODIC ACID  
100 ml 5% SILVER NITRATE  
100 ml 0.25M SODIUM BORATE  
1 PINT 3% SODIUM THIOSULFATE

1 PINT 3% METHENAMINE  
100 ml 0.2M BORIC ACID  
1 PINT 0.2% GOLD CHLORIDE  
1 PINT NUCLEAR FAST RED STAIN

**PRINCIPLE:** This stain kit demonstrates basement membrane and reticulum fibers.

**SPECIMEN:** Any well fixed paraffin embedded tissue cut at 5 microns; use strong tissue adhesive!

**QUALITY CONTROL:** American MasterTech Scientific Recommended Control Slide: Reticulum, CSRET25

**WORKING METHENAMINE-SILVER SOLUTION:** Prepare fresh solution just prior to use!

Prepare *Borate Buffer, pH 8.2* by adding 6.5 ml of **0.2 M BORIC ACID SOLUTION** to 3.5 ml of **0.25 M SODIUM BORATE SOLUTION**, mix thoroughly and set aside. In a separate container combine 42.5 ml of **3% METHENAMINE SOLUTION** and 2.5 ml of **5% SILVER NITRATE SOLUTION**, then add the 10 ml of *Borate Buffer, pH 8.2*, mix thoroughly and filter through medium filter paper into a chemically clean glass coplin jar or a suitable unused plastic staining container.

**PROCEDURE:**

1. Deparaffinize section using Xylene or Xylene Substitute and hydrate through alcohols as usual.
2. Rinse slide through several changes of Distilled water.
3. Place slide in **0.5% PERIODIC ACID** for 11 minutes.
4. Rinse slide thoroughly in Distilled water.
5. Place slide into a coplin jar or plastic container of fresh **Working Methenamine-Silver Solution**, then place container in a 65° to 70° C waterbath. Incubate slide for 60 to 75 minutes, checking slides microscopically every 10 minutes after the section becomes a medium brown color. NOTE: Check slide quickly! Do not allow slide to cool or uneven staining will occur; rinse slide in hot Distilled water before returning it to the **Working Methenamine-Silver Solution**. Remove slide from **Working Methenamine-Silver Solution** when basement membrane and reticulum fibers are intensely black and background tissue is dark yellow-brown.
6. Rinse slide through 3 changes of room temperature Distilled water.
7. Place slide in **0.2% GOLD CHLORIDE** for 1 minute; section should be gray-black.
8. Rinse slide through 2 changes of room temperature Distilled water.
9. Place slide in **3% SODIUM THIOSULFATE** for 1 to 2 minutes.
10. Rinse slide in running Tap water for 10 minutes.
11. Place slide in **NUCLEAR FAST RED STAIN** for 3 to 5 minutes.
12. Rinse slide through 3 changes of room temperature Distilled water.
13. Dehydrate slide through 3 changes of Absolute Alcohol.
14. Clear slide through 3 changes of fresh Xylene or Xylene Substitute.
15. Coverslip slide using a permanent mounting media.

**RESULTS:**

Basement membranes and reticulum fibers: **BLACK**  
Nuclei and Background: **PINK TO RED**

**REFERENCE:** Sheehan DC Hrapchak BB: Theory and Practice of Histotechnology; Second Edition; 186 -187, 1980.  
Luna LG: Histopathologic Methods and Color Atlas of Special Stain and Tissue Artifacts; American Histolabs, Inc.; 400 - 402, 1992.

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