Jones PAS-M Stain Kit

Periodic Acid Schiff Methenamine Silver Stain for Basement Membranes

DESCRIPTION
The Jones’ Basement Membrane Stain Kit is used to identify basement membranes, specifically glomerular and tubular basement membranes in renal tissue cut at 2 microns. Hematoxylin and Eosin stained sections of most tissues do not distinguish basement membranes very well; however, the basement membranes in the glomerulus are more conspicuous, especially in disorders such as membranous nephropathy or diabetes. These membranes are stained strongly positive using oxidation-aldehyde techniques, methenamine silver, etc., which are employed by the Jones’ PAS-M Stain Kit.

Note for Plastic Sections: Lay slides flat and allow to dry for several hours in a 60° oven. The bottom of the oven has a metal plate, this position should work well to keep the temperature constant while the slides are in the oven. The use of alcohols to dehydrate plastic sections causes them to wrinkle, partially or completely fall off the slide.

METHOD
1. Rehydrate sections to distilled water, use acid cleaned glassware and non metal forceps.
2. Oxidize with solution A at room temperature for 10 minutes for paraffin sections and 15 min for plastic sections.
3. Wash in 5 changes of distilled water for a few seconds.
4. Treat sections with solution B for 5 minutes
5. Wash in 5 changes of distilled water for a few seconds.

Preparation of Preheated Working Solution (Use solutions C,D & E)
Prepare working silver solution fresh each time, add in the following order in an acid cleaned coplin jar and do not use metal forceps

Solution C -------------- 1ml
Solution D ------------- 19ml
Solution E ------------- 2ml
Distilled water --------- 20 ml

6. Place slides in methenamine silver working solution and move slides and coplin jar to a water bath at that has been set 43°C, already preheated to 43°C for 2 minutes
7. Transfer to a 50°C waterbath for 15 minutes, rinse in distilled water and check with a microscope, the basement membranes should be dark brown. If they are light return to the methenamine silver solution for another 1-2 minutes, Check slides again, slides stain quickly do not overstain the basement membrane.
8. Rinse well in distilled water, 5 changes
9. Tone sections with solution F for 1 minute
10. Rinse in 2 changes of distilled water
11. Treat sections with solution G for 2 minutes
12. Wash with running tap water for 5 minutes
13. Counterstain in solution H for 5 seconds
14. Rinse excess NFR green off slide with distilled water, 3 changes
15. Dehydrate, clear and mount paraffin sections, plastic sections must be air dried and then dipped in xylene and mount.

SAFETY
Work in a well ventilated area, wear protective clothing, gloves and safety glasses when preparing reagents.

STORAGE AND HANDLING
Storage - Store at room temperature
Handling - Glove, chemical goggles & fume hood

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