ScyTek LABORATORIES INC.

Instructions For Use

JSK-IFU

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Jones Methenamine Silver Stain Kit

(For Basement Membrane)

Description and Principle

The Jones Stain Kit is intended for use in histological demonstration of the basement membrane and reticular fibers. This procedure is ideal for staining renal glomerular basement membranes. The main function of the basement membrane and reticular fibers is to provide anchorage and support. They are normally found throughout the body, particularly in the kidney, spleen, and lung.

Basement membranes and reticular fibers are demonstrated by methenamine silver through oxidation of carbohydrates to aldehydes. The aldehydes formed by oxidation bind silver ions from the methenamine silver complex and reduce the silver to its metallic form.

Expected Results

Basement Membrane:	Black
Oxidizable Carbohydrates:	Black
Nuclei:	Red
Cytoplasm:	Light Pink

Kit Contents

	Storage
1. Periodic Acid Solution	2-8°C
2. Methenamine Solution	2-8°C
3. Silver Nitrate Solution (5%)	2-8°C
4. Borax Solution	18-25°C
5. Gold Chloride Solution (0.2%)	2-8°C
6. Sodium Thiosulfate Solution (5%)	18-25°C
7. Nuclear Fast Red Solution (Enhanced)	18-25°C

Suggested Controls *(not provided)*

Kidney cut at 2 microns, Lung, Spleen

Uses/Limitations

For In-Vitro Diagnostic use only.
Do not use if reagents become cloudy or precipitate
Do not use past expiration date.
Use caution when handling reagents.
Non-Sterile
Intended for FFPE sections cut at 5-10µm.
This procedure has not been optimized for frozen sections.
Frozen sections may require protocol modification.

Storage

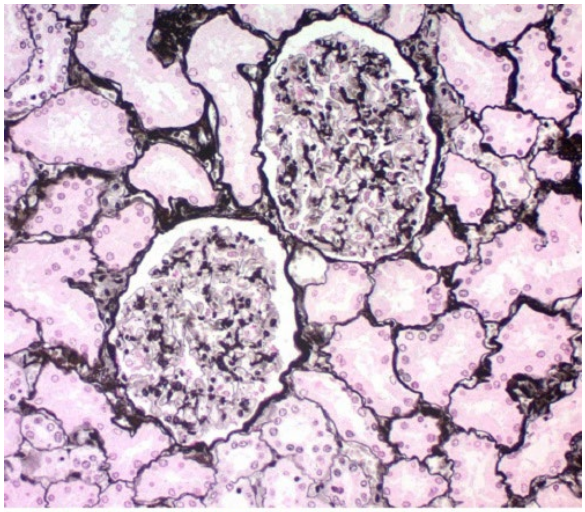
Store kit contents according to individual bottle labels

Safety and Precautions

Please see current Safety Data Sheets (SDS) for this product and components GHS classification, pictograms, and full hazard/precautionary statements.

Important Notes

1. All glassware used in this procedure should be chemically cleaned and rinsed thoroughly in distilled water.
2. Do not use metal forceps to remove slides from reagents. Use plastic forceps only.
3. Equilibrate all reagents to room temperature prior to use.
4. Increasing the number of slides present in working Methenamine Silver Solution will decrease the rate of silver staining. Monitor slides during Methenamine Silver incubation to achieve desired intensity.



Normal Kidney glomerular basement membrane cut at 2 micron thickness. Stained for 1 hour with working Silver Methenamine Solution. Toned for 30 seconds with Gold Chloride Solution (0.2%). Magnification 20X.

Procedure

1. Deparaffinize sections and hydrate to distilled water.
2. Incubate slide in Periodic Acid Solution for 15 minutes.
3. Rinse in 2 changes of distilled water.
4. Combine the following for a working Methenamine Silver Solution:

   42 ml Methenamine Solution
   2.5 ml Silver Nitrate Solution (5%)
   6 ml Borax Solution

   Note: Mixed solution may not be stored for reuse.

5. Place working Methenamine Silver Solution in 65° centigrade water bath and allow temperature to equilibrate.
6. Incubate slide in working Methenamine Silver Solution for 20-60 minutes. Dip slide in "hot" distilled water and check under a microscope for evaluation of silver impregnation. Basement membrane and reticular fibers should be black. If color is not sufficient, return the slide to working Methenamine Silver Solution checking frequently until desired staining intensity is achieved.
7. Rinse in 4 changes of distilled water.
8. Tone slide in Gold Chloride Solution (0.2%) for 30 seconds.
9. Rinse in 4 changes of distilled water.
10. Incubate slide in Sodium Thiosulfate Solution (5%) for 2 minutes.
11. Rinse in tap water followed by 2 changes of distilled water.
12 Incubate slide in Nuclear Fast Red Solution for 2 minutes.
13. Rinse slide quickly in distilled water.
14. Rinse slide using absolute alcohol.
15. Dehydrate in 3 changes of absolute alcohol, clear, and mount in synthetic resin.

References

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