

Methenamin Silver Method for Argentaffin Cells

Catalog #: 26481-Series

Fixation:

Formalin, Bouin's (#15990-01) or other Formalin containing fixatives. Dichromates and mercury salts impair contrast between granules and background.

Sections:

Cut paraffin at 3 - 5µm sections.

Staining Procedures:

1. Deparaffinize and hydrate slides to distilled water.
2. Treat for 10 minutes in Weigert's Iodine (#26481-01) followed by bleaching for 10 minutes in Sodium Thiosulfate, 5% (#26481-02). Wash for 10 minutes in running water. Rinse in two changes in distilled water.
3. Place section in chemically clean coplin jars containing the buffered methenamine-silver nitrate solution at room temperature and place in 60°C paraffin oven for 3 – 3 ½ hours. (preheating the solution to 60°C reduces impregnation time by ½ to 1 hour). Rinse in distilled water.
 - a. * prepare methenamine-silver nitrate by mixing:
Silver Nitrate, 5% (#26481-03) - 5 ml
Methenamine, 3% (#26481-04) - 100 ml
For working buffered stain add 8 ml of Borate Buffer, pH 7.8 (#26481-05) to 30 ml of methenamine-silver nitrate. Use chemically clean glassware throughout.
4. Tone for 10 minutes in Gold Chloride, 0.1% (#26481-06). Rinse in distilled water.
5. Fix in Sodium Thiosulfate, 5% (#26481-02) for 2 minutes. Wash in running water for 5 minutes
6. Counterstain in Acetic Safranin O, 0.1% (#26481-07) for 5 minutes.
7. Dehydrate with Acetone, clear in Xylene and mount.

Stain Results:

Argentaffin Cells	Black
Coarse connective tissue of the submucosa	Variable amount of blackening by 3 – 3 1/2 hour of impregnation
Granules of eosinophil leukocytes, nuclei, smooth muscle & surface epithelium	Show additional blackening after incubation beyond 3 – 3 ½ hour.
Granules of mast cells	Remain red after nuclei & reticulum are blackened

References:

- Clark, G, ed.: Staining Procedures, 3rd ed.: Baltimore: Williams & Wilkins Co., 1973, p. 161
- Gomori, G.: Arch Path., 45:48, 1948
- Burtner, H.J. and Lillie, R.D., Stain Tech., 24:225-7, 1949.