A-122 RAPID GROCOTT’S METHENAMINE-SILVER NITRATE METHOD FOR FUNGI (GMS) AND PNEUMOCYSTIS CARINII
(MODIFIED FOR MICROWAVE)

**FIXATION:** Formalin, 10% Buffered Neutral
**SECTION:** Cut paraffin section at 4-6 microns

**STAINING PROCEDURE:**

1. Deparaffinize and hydrate to distilled water.
2. Oxidize in Chromic Acid, 10% (A-122-1), and microwave for 20 seconds at maximum power.
3. Let stand for 10 seconds in hot solution, then rinse in running tap water.
4. Place in Sodium Bisulfite, 1% Aq. (A-122-4), for 60 seconds.
5. Rinse in running tap water followed by 3 changes of distilled water.
6. Place slides in Methenamine Silver Nitrate Working Solution and microwave for 20 seconds at maximum power.

*To prepare working solution:

**A. Methenamine – Silver Nitrate Stock Solution**
- Silver Nitrate, 5%, Aq. (A-122-3A) – 5ml
- Methenamine, 3%, Aq. (A-122-3B) – 100ml

**B. Methenamine Silver Nitrate Stock Working Solution**
- Methenamine-Silver Nitrate Stock Sol’n Stock 25 ml
- Distilled Water 25 ml
- Sodium Borate 5%, Aq. (A-122-2) 2 ml

Prepare fresh prior to use. Do not use if cloudy.

7. Let slides stand in hot solution which has been placed in a beaker of water at 60°C for 30 or more seconds. The slides should remain until they are pale brown.
8. Rinse slides in 3 changes of distilled water.
9. Tone in Gold Chloride 1%, aq. (A-122-5) for 10 seconds.
10. Rinse slides in distilled water.
11. Place in Sodium Thiosulfate, 2% Aq. (A-122-6) for 1 minute.
12. Wash in running tap water for 2 minutes.
13. Place in Light Green Stock Solution, 0.2% (A-122-7), for 30 seconds.
14. Rinse in distilled water.
15. Dehydrate, clear and mount in resinous medium.

**RESULTS:**

- Fungi ……………………………………. black, sharp detail
- Pneumocystis carinii ……………………. black
- Mucin…………………………………. dark gray - taupe
- Inner areas Mycelia, Hyphae……………. rose
- Background……………………………… green, pale

**REFERENCES:**


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