TRICROME - MICROWAVE PROCEDURE

PURPOSE: This technique is a variation of the Masson's Trichrome Stain utilizing the microwave oven.

TECHNIQUE: Cut paraffin sections at 4m

EQUIPMENT & SUPPLIES: Microwave oven, plastic coplin jars, rinse all glassware in DI water.

REAGENTS:

Weigert's Iodine:
- Potassium iodide 2.0 gm
- Iodine crystals 1.0 gm
- Distilled water 100.0 ml

Dissolve potassium iodide in approximately 10 ml of the distilled water, then add the iodine crystals, dissolve. Add the remaining distilled water. Stable for 1 year.

Gills III Hematoxylin:
- Purchased through Baxter

Biebrich Scarlet
- See Masson’s Trichrome

Phosphotungstic / Phosphomolybdic Acid:
- See Masson’s Trichrome

Alcian Blue:
- See Masson’s Trichrome

5% Hypo:
- See Stock Solutions

1% Acetic Acid:
- See Masson’s Trichrome

SAFETY: Use the exhaust hood when irradiating solutions, keep hot, uncapped, solutions under the hood. Wear gloves, goggles and lab coat. Avoid contact and inhalation of dyes and chemicals.

Iodine: heating will generate copious amounts of toxic fumes. Dermal sensitizer, irritant to eyes, skin and respiratory system. Corrosive.

Phosphomolybdic, phosphotungsic acid powders, and acetic acid solutions are skin and eye irritants, and strong corrosives.
CONNECTIVE TISSUE

MICROWAVE TRICHOME

Sodium thiosulfate: Toxic on ingestion. Can irritate the stomach. Irritant to skin, eyes and respiratory tract.

PROCEDURE:
1. Deparaffinize and hydrate to distilled water.
2. Mordant in Weigert's iodine for 45 seconds, let stand 1 minute.
3. Wash in water.
4. Hypo solution 20 dips or until iodine is removed.
5. Rinse in distilled water.
6. Gill's III Hematoxylin, microwave 45 seconds.
7. Wash in tap water until blue, rinse in distilled water.
8. Biebrich scarlet, microwave 45 seconds.
9. Distilled water.
10. Phosphotungstic-phosphomolybdic acid, microwave 45 seconds.
11. Quick 1 dip in distilled water.
12. Aniline blue, microwave Hi power, 20 seconds.
13. Distilled water.
14. 1% acetic acid rinse for 30 seconds.
15. Dehydrate, clear and mount.

RESULTS:
Nuclei blue-black
Cytoplasm, muscle, and erythrocytes red
Collagen blue

REFERENCES:
Sigma Microwave Procedures, Brinn, N
Crookham, J, Dapson, R, Hazardous Chemicals in the Histopathology Laboratory, 2nd ED, 1991, Anatech
PROCEDURE CARD
TRICHROME, MICROWAVE PROCEDURE - COLLAGEN

PROCEDURE:
1. Deparaffinize and hydrate to distilled water.
2. Mordant in Weigert's iodine for 45 seconds, let stand 1 minute.
3. Wash in water.
4. Hypo solution 20 dips or until iodine is removed.
5. Rinse in distilled water.
6. Gill's III Hematoxylin, microwave 45 seconds.
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8. Biebrich scarlet, microwave 45 seconds.
9. Distilled water.
10. Phosphotungstic-phosphomolybdic acid, microwave 45 seconds.
11. Quick 1 dip in distilled water.
12. Aniline blue, microwave Hi power, 20 seconds.
13. Distilled water.
14. 1% acetic acid rinse for 30 seconds.
15. Dehydrate, clear and mount.

RESULTS:
Cytoplasm, muscle, nuclei blue-black
Erythrocytes red
Collagen blue

SAFETY/ PPE: Use the exhaust hood when irradiating solutions, keep open staining jars of hot solutions under the hood.

Weigert's Iodine:
Potassium iodide 2.0 gm
Iodine crystals 1.0 gm
Distilled water 100.0 ml
Dissolve potassium iodide in approximately 10 mls of the distilled water, then add the iodine crystals, dissolve. Add the remaining distilled water. Stable for 1 year.

5% Hypo:
See Stock Solutions

Gills III Hematoxylin:
Purchased through Baxter

Masson's Trichrome Staining Solutions:
See Masson's Trichrome
Weigert's Iodine:
Potassium iodide 2.0 gm
Iodine crystals 1.0 gm
Distilled water 100.0 ml

Dissolve potassium iodide in approximately 10 mls of the distilled water, then add the iodine crystals, dissolve. Add the remaining distilled water. Stable for 1 year.

CAUTION: Dermal sensitizer, irritant to eyes, skin, and respiratory system.

DATE:__________________________
TECH:__________________________
EXPIRATION:_____________________

Weigert's Iodine
DATE:__________________________
TECH:__________________________

Aniline Blue
DATE:__________________________
TECH:__________________________

Biébrich Scarlet
DATE:__________________________
TECH:__________________________

Gill's III Hematoxylin
DATE:__________________________
TECH:__________________________

Phosphotungstic / Phosphomolybdic Acid
DATE:__________________________
TECH:__________________________