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**TRICHROME - MICROWAVE PROCEDURE**

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**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.

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

**5% Hypo:**

See Stock Solutions

**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

**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, phosphotungstic acid powders, and acetic acid solutions are skin and eye irritants, and strong corrosives.

CONNECTIVE TISSUE

**MICROWAVE TRICHROME**

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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

Prepared: \_\_\_\_\_ By: \_\_\_\_\_

Approved: \_\_\_\_\_ By: \_\_\_\_\_

## PROCEDURE CARD

### TRICHROME, MICROWAVE PROCEDURE - COLLAGEN

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#### PROCEDURE:

1. Deparaffinize and hydrate to distilled water.
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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

#### Gill's 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: \_\_\_\_\_

**WEIGERTS IODINE**

DATE: \_\_\_\_\_

TECH: \_\_\_\_\_

**ANILINE BLUE**

DATE: \_\_\_\_\_

TECH: \_\_\_\_\_

**BIEBRICH SCARLET**

DATE: \_\_\_\_\_

TECH: \_\_\_\_\_

**GILL'S III HEMATOXYLIN**

DATE: \_\_\_\_\_

TECH: \_\_\_\_\_

**PHOPHOTUNGSTIC /  
PHOSPHOMOLYBDIC ACID**

DATE: \_\_\_\_\_

TECH: \_\_\_\_\_