

## Masson's Trichrome for Connective Tissue

**Catalog #:** 26367-Series

### Fixation:

Bouin's (#15990-01) or 10% Buffered Formalin (#15741-01)

### Sections:

Paraffin at 6µm

### Staining Procedures:

1. Deparaffinize and hydrate to distilled water.
2. Mordant in Bouin's Fixative (#26367-01) for 1 hour at 56°C or overnight at room temperature.
3. Cool and wash in running water until yellow color disappears.
4. Rinse in distilled water.
5. Stain in Weigert's Iron Hematoxylin Working\* solution for 5 minutes. Wash in running water for 10 minutes.
  - a. To prepare working solution, mix equal parts of Weigert's Iron Hematoxylin A (#26367-02) and Weigert's Iron Hematoxylin B (#26367-03).
6. Rinse in distilled water.
7. Place in Biebrich Scarlet-Acid Fuchsin (#26367-04) for 15 minutes.
8. Rinse in distilled water.
9. Place in Phosphomolybdic Acid-Phosphotungstic Acid (#26367-05) for 15 minutes.
10. Stain in Aniline Blue Solution (#26367-06) for 10 to 20 minutes.
11. Rinse in distilled water.
12. Differentiate in Acetic Acid, 1% (#26367-07) for 3 to 5 minutes. Discard solution.
13. Dehydrate in 95% alcohol and absolute alcohol, and clear in xylene, 2 changes each.
14. Mount.

Shorten staining times when using 2 – 4 microns sections of renal biopsy specimens:

Biebrich Scarlet – Acid Fuchsin: 5 minutes

Phosphomolybdic Acid-Phosphotungstic Acid: 10-12 minutes

Aniline Blue Solution: 3 minutes

### Stain Results:

Nuclei	Black
Cytoplasm, Keratin, Muscle Fibers	Red
Collagen, Mucin	Blue

NOTE: To ensure proper mordanting Bouin's Fixative should be preheated to 58 – 60°C

### References:

Sheehan and Hrapchak. Theory and Practice of Histotechnology. St. Louis. The Mosby Comp. 1980, p. 190