

## **F-367      MASSON'S TRICHOME FOR CONNECTIVE TISSUE**

**Fixation:** Bouin's Fixative (F-40) or 10% Buffered Formalin (F-113)

**Sections:** Paraffin cut at 6 micrometers.

**PROCEDURE:**

1. Deparaffinize and hydrate to distilled water.
2. Mordant in Bouin's Fixative (F-367-1) 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. Place in Weigert's Iron Hematoxylin Solution for 10 minutes.

\*\* To prepare Weigert's Iron Hematoxylin Working Solution:

Mix equal parts of Weigert's A (F-367-2A)  
with equal parts of Weigert's B (F-367-2B)

6. Wash in running tap water for 10 minutes.
7. Rinse in distilled water.
8. Place in Biebrich Scarlet-Acid Fuchsin (F-367-3) for 15 minutes.
9. Rinse in distilled water.
10. Place in Phosphotungstic-Phosphomolybdic Acid (F-367-4) for 10 to 15 minutes.
11. Place in Aniline Blue Solution (F-367-5) for 10-20 minutes.
12. Rinse in distilled water.
13. Place in Acetic Acid, 1% (F-367-6) for 3 to 5 minutes. Discard solution.
14. Dehydrate in 95% Alcohol, Absolute Alcohol, and clear in Xylene, 2 changes each.
15. Mount.

Shorten staining times when using 2-4 micron sections of renal biopsy specimens:

BiebrichScarlet- 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 insure proper mordanting. Bouins Fixative should be preheated to 58-60°C.

**References:**

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

