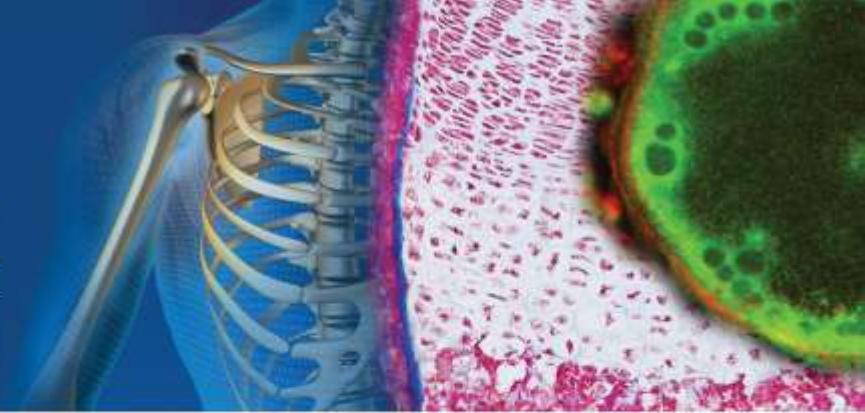


# CENTER *for* MUSCULOSKELETAL RESEARCH



## MASSON TRICROME STAIN

**Paraffin-embedded, formalin fixed tissue**

Notes: Muscle fibers, keratin ----- red  
Nuclei ----- black  
Collagen ----- blue

SOLUTIONS:

### Bouin's Solution

Bouin's solution – Fisher Scientific: NC0827069

### Modified Weigert's Iron Hematoxylin

Solution A

Hematoxylin crystals, C.I. 75290 -----	4.0 g
Alcohol, 80% -----	200 ml

Solution B

Ferric chloride (EMS 15510) -----	8.0 g
Distilled water -----	190 g
HCl, concentrated 36.5-38% -----	2.0 ml

### Working Modified Weigert's Iron Hematoxylin

Equal parts of Solution A and Solution B mixed well, keep in dark.

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## MASSON TRICROME STAIN

### Biebrich Scarlet-Acid Fuchsin Solution

Biebrich scarlet, C.I. 26905 -----	2.25 gm
Acid fuchsin, C.I. 42685 -----	0.25gm
Acetic acid, glacial -----	2.5 gm
Distilled water -----	250 ml

### 1% Phosphomolybdic Acid Solution

Phosphomolybdic acid -----	2.5 gm
Distilled water -----	250 ml

### 1.8 % Aniline Blue Solution

Aniline blue, C.I. 42755 -----	4.5 gm
Acetic acid, glacial -----	4.5 ml
Distilled water -----	250 ml

### 1% Acetic Acid Solution

Acetic acid, glacial -----	2.5 ml
Distilled water -----	250 ml

### Acid-Alcohol Differentiation Solution

0.5% hydrochloric acid in 70% alcohol

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## MASSON TRICROME STAIN

### STAINING PROCEDURE:

1. Deparaffinize and hydrate to distilled water
2. Mordant in Bouins's solution which has been preheated to 58° C for **15 minutes**
3. Wash slides with distilled water
4. Modified Weigert's Iron Hematoxylin (keep in dark) for **5 minutes**
5. Wash with **3-4 changes** of distilled water
6. Differentiate in acid-alcohol solution for **3-5 seconds**
7. Wash slides with **3-4 changes** of distilled water
8. Biebrich Scarlet-Acid Fuchsin solution for **5 minutes**
9. Rinse in **3 changes** of distilled water
10. 1% Phosphomolybdic acid solution for **2 minutes**
11. Aniline Blue solution for **5 minutes**
12. Rinse with **1 change** of distilled water
13. Acetic acid solution for **30 seconds**
14. Rinse with **2 changes** of distilled water
15. Dehydrate in graded alcohols
16. Clear in Xylene
17. Coverslip

2/2020