Usage procedure for:

Martius Scarlet Blue (MSB) Stain Kit

Avantik Kit Item #: RS4607-125 | RS4607-250 | RS4607-500

Solutions Provided in Stain Kit:
Iron Hematoxylin, Solution A
Iron Hematoxylin, Solution B
Martius Yellow (MSB) Stain
Crystal Scarlet (MSB) Stain
Phosphotungstic Acid, 1%
Aniline Blue (MSB) Stain

Working solution preparation (prepare just before use):

**Working Iron Hematoxylin Solution**
Iron Hematoxylin, Solution A: 25mL
Iron Hematoxylin, Solution B: 25mL
*Final volume may be adjusted as long as equal parts Solution A and Solution B are used.

Conventional Procedure

1. Deparaffinize and hydrate sections through alcohol to distilled water.
2. **OPTIONAL:** If specimen was not originally fixed in Bouin’s Solution, post fix by placing sections in Bouin’s Solution overnight at room temperature or 1 hour at 56°C. Formaldehyde fixation is acceptable, however, the use of Bouin’s Solution will enhance staining. 
   If this step is omitted, proceed directly to Step 4.
3. After slides have cooled, thoroughly rinse sections in running tap water until excess Bouin’s has been removed (5-10 minutes).
5. Wash sections in running tap water for 5-10 minutes.
6. Stain in Martius Yellow (MSB) Stain for 2-3 minutes.
7. Wash sections well in water.
8. Stain in Crystal Scarlet (MSB) Stain for 5-10 minutes.
9. Wash section well in water.
10. Treat sections with Phosphotungstic Acid, 1% checking microscopically until only the fibrin remains red. Collagen must be decolorized. **This process may take up to 10 minutes or longer.**
11. Wash sections well in water.
12. Stain in Aniline Blue (MSB) Stain for 2-5 minutes. Check microscopically until collagen is the desired intensity of blue.
13. Dehydrate rapidly in three changes of 100% Alcohol.
14. Clear and mount with appropriate mounting medium.
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Results:
Fibrin: Red (fresh fibrin may appear yellow and old fibrin blue)
Muscle: Pink to Pale Red
Erythrocytes: Yellow
Nuclei: Blue-Black
Collagen: Blue
Other Connective Tissue: Blue

Kit Components:

Avantik Item Numbers:
Iron Hematoxylin, Solution A: RS4418-250 (250 mL) | RS4418-500 (500 mL)
Iron Hematoxylin, Solution B: RS4419-250 (250 mL) | RS4419-500 (500 mL)
Martius Yellow (MSB) Stain: RS4464-250 (250 mL) | RS4464-500 (500 mL)
Crystal Scarlet (MSB) Stain: N/A
Phosphotungstic Acid, 1%: RS4436-250 (250 mL) | RS4436-500 (500 mL)
Aniline Blue (MSB) Stain: RS4592-250 (250 mL) | RS4592-500 (500 mL)