
F-384 MOVAT'S PENTACHROME METHOD

FIXATION: Formalin, 10% Buffered Neutral

SECTION: Paraffin, 6 microns

STAINING PROCEDURE:

1. Deparaffinize and hydrate to distilled water, then rinse thoroughly in distilled water.
2. Stain in **Alcian Blue**, (F-384-2) 20 min. Wash in running water, 3 min.
3. Tone in **Alkaline Alcohol** (F-384-6), 2 hrs. (This converts the Alcian Blue into the insoluble pigment monastral fast blue).
4. Wash in running water, 10 min. and then rinse in 70% alcohol.
5. Stain in **Resorcin-Fuchsin Working**, 16 hrs. Wash in running water, 10 min. and then in distilled water.

*To prepare working solution mix:

Resorcin Fuchsin Stock (F-384-3) 10 ml
70% Alcohol 100 ml
Hydrochloric Acid, conc. 2 ml

6. Stain in **Working Weigert's Hematoxylin** [(mix 1 volume of **Weigert's A** (F-384-1A) plus 1 volume of **Weigert's B** (F-384-1B) for 15 minutes. Rinse in running water and then in distilled water.
7. Stain in **Woodstain Scarlet – Acid Fuchsin**, (F-384-4), 5 min. Rinse in **Acetic Acid, 0.5%** (F-384-8).
8. Differentiate in **Phosphotungstic Acid, 5%**, (F-384-9), 10-20 min. Check microscopically until the background returns to blue and the collagen is pale pink.
9. Rinse in **Acetic Acid, 0.5%** (F-384-8). Take the slide through three changes of absolute alcohol of **high purity**.
10. Stain in **Alcoholic Safran Solution**, (F-384-5), for 5-15 minutes.
11. Dehydrate through three changes of 100% alcohol. Clear in three changes of Xylene, high purity. Check with a microscope; the collagen should be yellow, if not return to the safran stain.
12. Mount with Permount (M-18).

STAIN:

Black	Nuclei
Purple-Black	Elastic Fibers
Dark Red-Red	Fibrinoid, Muscle
Blue-Blue Green	Ground or Background substance
Yellow	Collagen, Reticulum Fibers

REFERENCES:

Movat, H.Z.: Arch. Path.; 60: 289 (1955).

Luna, L.G., ed.: Manual of Histologic Staining Methods; 3rd. ed., McGraw-Hill Book Co., c. 1968, p. 95.

