medilines

Masson's Trichrome

for Collagen and Muscle

DESIGN TO DEMONSTRATE

Used to differentiate between collagen and smooth muscle in tumors, and the increase of collagen in diseases such as cirrhosis. Routine stain for liver and kidney biopsies.

REAGENTS & PREPARATIONS

Masson Solution A	
Masson Solution B	
Masson Solution 1:	Mix A and B in equal volume (Solution will remain stable for one weak. Preferably prepare fresh)
Decolourizer:	Ready to use
Masson Solution 2:	Ready to use
Masson Solution 3:	Ready to use
Masson Solution 4:	Ready to use

TISSUE SAMPLE

4-5µ paraffin sections of 10% neutral buffered formalin.
Trichrome stains often benefit from initial Bouin, formal sublimate or B5 fixation. Zenker and Helly fixation is usually satisfactory.

Note: Formalin variants are generally sufficient, but staining can be intensified by secondary fixation (Mordant) in Bouin's Fluid

Place section after the 1st step of following staining procedure in Bouin's fluid for an hour at 56-60°C (or microwave 1 minute, allow to stand for 15 minutes} and wash in running tap water for 5 minutes to remove Picric Acid.

STAINING PROCEDURES

1. Remove Paraffin through xylene, rehydrate through graded ethanol and bring sections to distilled water just prior to staining.
2. Place into **Masson Solution 1** for 5-7 minutes
3. Place in **Decolourizer** for 5 seconds. Wash in running tap water for 30 seconds and rinse in distilled water.
4. Place into **Masson Solution 2** for 5-10 minutes.
5. Wash with distilled water.
6. Place into **Masson Solution 3** for 5-10 minutes.
7. Rinse in distilled water.
8. Place into **Masson Solution 4** for 5-10 minutes.
9. Rinse well with distilled water
10. Dehydrate up through graded ethanol to absolute ethanol.
11. Clear in xylene and mount with a resinous medium.

Expected results

- Muscle, Cytoplasm, Erythrocytes – Red
- Collagen – Green
- Nuclei – Dark brown

Positive Controls

Skin, Lung, Stomach, Intestine.

Notes

1. Overnight formalin fixation is usually satisfactory, but avoid rapid fixation with formalin and short processing, as this produces tissues that stain poorly even with the secondary fixation specified
2. Once a satisfactory stain has been achieved, the time should remain constant. Any change in fixation, processing or section thickness warrants reviewing the staining time.
3. Avoid microbial contamination of reagents; otherwise increase in nonspecific staining may occur.
4. The user must validate any changes made to the factory-released procedure.
5. Store reagent packs in an upright position when not in use.
6. Keep reagent packs out of direct sunlight and away from heat-generating sources.
7. Wear appropriate Personal Protective Equipment to avoid contact with eyes and skin.

Reference

Theory and practice of histological techniques,
Bancroft, J. D. and Stevens, A.
Churchill Livingstone, London, England

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