

# VitroView<sup>™</sup> Masson's Trichrome Stain Kit

# Cat. No. VB-3016

#### Introduction

Masson's Trichrome Stain Kit is used to differentiate between collagen and smooth muscle in tumors, and the increase of collagen in diseases such as cirrhosis. It is a routine stain for liver and kidney biopsies. This kit can be used for FFPE and frozen tissue slides.

# **Kit Components**

- 1. Bouin's solution -----250 ml
- Weigert's Hematoxylin Solution A -----125 ml
  Weigert's Hematoxylin Solution B ------125 ml
- Annue Blue Solution -----250 ml
  Biebrich Scarlet-Acid Fuchsin Solution ------250 ml
- Biebrich Scallet-Acid Fuchsin Solution ------250 mi
  Phosphomolybdic-Phosphotungstic Acid Solution ------250 mi
- 7. 1% Acetic Acid Solution -----250 ml

# Storage

Room temperature.

# Protocol

#### For formalin-fixed, paraffin-embedded (FFPE) tissue sections:

- 1. Deparaffinize in Xylene I for 6 minutes and II for 6 minutes.
- 2. Rehydrate.
  - a. Ethanol 100% (2 minutes)
  - b. Ethanol 100% (2 minutes)
  - c. Ethanol 95% (2 minutes)
  - d. Ethanol 95% (2 minutes)
  - e. Ethanol 70% (2 minutes)
- 3. Rinse in distilled water (5 minutes).
- 4. Mordant in Bouin's Solution, 60°C for 1 hour.
- 5. Wash in running tap water for 5 minutes to remove the picric acid.
- 6. Prepare Weigert's working hematoxylin: mix Weigert's Hematoxylin Solution A and B at 1:1 ratio.
- 7. Weigert's working hematoxylin for 10 minutes. *Note: Discard after use.*
- 8. Blue in running tap water for 5 minutes, rinse once for 1 min in distilled water.
- 9. Biebrich Scarlet-Acid Fuchsin Solution for 5 minutes.
- Note: Solution may be used twice only, then discarded.
- 10. Rinse once for 1 min in distilled water.
- 11. Phosphotungstic/phosphomolybdic acid solution for 10 minutes, discard solution.
- 12. Transfer directly into Aniline Blue Solution for 10 minutes.
- 13. Rinse once for 1 min in distilled water.
- 14. Acetic Acid Solution for 1 minute, discard solution.
- 15. Wash slides,  $2 \min \times 2 \inf dH_2O$ .
- 16. Dehydrate with 2 changes of 95% Ethanol and 2 changes of 100% Ethanol (2 minute per change).

17. Clear with 3 changes of xylene (5 minutes per change) and coverslip with Permount or other suitable organic mounting medium.

#### For Frozen Sections:

- 1. Fix frozen sections in 10% formalin (not Zinc) for 30 minutes.
- 2. Rinses in distilled water 3×3min.
- 3. Mordant in Bouin's Solution, 60°C for 1 hour.
- 4. Wash in running tap water for 5 minutes to remove the picric acid.5. Weigert's working hematoxylin for 7 minutes.
- Note: Discard after use.
- 6. Blue in running tap water for 5 minutes, rinse in distilled water.
- 7. Biebrich Scarlet-Acid Fuchsin Solution for 5 minutes.
- Note: Solution may be used twice only, then discarded.
- 8. Rinse in distilled water.
- 9. Phosphotungstic/phosphomolybdic Acid Solution for 10 minutes, discard solution,
- 10. Transfer directly into Aniline Blue Solution for 5 minutes.
- 11. Rinse in distilled water.
- 12. 1% Acetic acid for 1 minute, discard solution.
- 13. Wash slides,  $2 \min \times 2 \inf dH_2O$ .
- 14. Dehydrate with 2 changes of 95% Ethanol and 2 changes of 100% Ethanol (2 minute per change).
- 15. Clear with 3 changes of xylene (5 minutes per change) and coverslip with Permount or other suitable organic mounting medium.

# **Expected results**

Cytoplasm, keratin, muscle fibers, Erythrocytes	red
Nuclei	
Collagen and mucus	blue

#### Note

This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

#### Precautions

Handle with care. Avoid contact with eyes, skin and clothing. Do not ingest. Wear gloves.

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