Modified Masson'S Trichrome Staining Solution

Cat #: A-CSH274

Size: 8×50mL

Storage: Store in dark at 2~8°C (12 months)

Product Description

In a narrow sense, connective tissue refers to the three types of fibers it contains: collagen fibers, reticular fibers, and elastic fibers, with collagen fibers being the most widely distributed and abundant. Masson's trichrome staining, also known as Masson's staining, is one of the most classic methods for connective tissue staining and is an authoritative and classic technique for staining collagen fibers. The term "trichrome staining" usually refers to staining the cell nucleus and selectively displaying collagen fibers and muscle fibers. The staining principle of this method is related to the size of the anionic dye molecules and tissue permeability: smaller molecules can easily penetrate dense and low-permeability tissues, while larger molecules can only enter loosely structured and highly permeable tissues. However, aniline blue has large molecular weight, so after Masson staining, muscle fibers appear red, and collagen fibers appear green (light green) or blue (aniline blue), primarily used to differentiate collagen fibers and muscle fibers.

The Modified Masson'S Trichrome Staining Solution differs from the conventional Masson's trichrome staining in the use of light staining of cell nucleus with Celestine Blue Hematoxylin. Its characteristics include short differentiation time (1-2 minutes), clear and vibrant colors, wide applicability, suitable for staining tissue sections in paraffin and frozen sections. And the stained sections have long preservation time and are not prone to fading. Collagen fibers appear blue, while muscle fibers, cell cytoplasm, cellulose, keratin, and red blood cells appear red, and cell nucleus appear blue-brown. It is mainly used to differentiate collagen fibers and muscle fibers.

Materials Supplied and Storage Conditions

Components	Size (8×50mL)	Storage conditions
Reagent A: Bouin Solution	50 mL	10~30°C
Reagent B: Celestine Blue Staining Solution	50 mL	Store in dark at 2~8°C
Reagent C: Mayer Hematoxylin Staining Solution	50 mL	Store in dark at 2~8°C
Reagent D: Acid Alcohol Differentiation Solution	50 mL	10~30°C
Reagent E: Ponceau S Fuchsin Staining Solution	50 mL	Store in dark at 10~30°C
Reagent F: Phosphomolybdic Acid Hydrate Solution	50 mL	Store in dark at 10~30°C
Reagent G: Aniline Blue Stain Solution	50 mL	Store in dark at 10~30°C
Reagent H: Weak Acid Solution	50 mL	10~30°C

Procedure

1. The tissues were fixed in 10% formalin fixative, followed by routine dehydration and embedding. The sections were cut to a thickness of 4 μm and then subjected to routine deparaffinization until water.

2. The sections were placed in Bouin Solution and incubated at room temperature overnight or placed in a 37°C incubator for 2 hours for mordanting. Then, the sections were rinsed with running water until the yellow color on the sections disappeared.

3. The sections were stained with Celestine Blue Staining Solution for 2-3 minutes, followed by a brief water rinse.

4. The sections were stained with Mayer Hematoxylin Staining Solution for 2-3 minutes, followed by a brief water rinse.

5. The sections were differentiated in Acid Alcohol Differentiation Solution for a few seconds, followed by a 10-minute water rinse.

6. The sections were stained with Ponceau S Fuchsin Staining Solution for 10 minutes, followed by a distilled water rinse.

7. The sections were treated with Phosphomolybdic Acid Hydrate Solution for approximately 10 minutes.

8. The supernatant was poured off, and without water rinsing, the sections were directly stained with Aniline Blue Stain Solution for 5 minutes.

9. The sections were treated with Weak Acid Solution for 2 minutes.

10. Rapid dehydration was performed using 95% ethanol. The sections were dehydrated in 100% ethanol three times, each time for 5-10 seconds.

11. The sections were cleared in xylene three times, each time for 1-2 minutes. Finally, neutral mounting medium was applied for sealing.

Staining Results

Collagen fiber	Blue
Muscle fibers, cell cytoplasm, cellulose, keratin, and red blood cell	Red
Cell nucleus	Blue-brown

Notes

1. Deparaffinization of the sections should be done as thoroughly as possible. The differentiation time of acid alcohol should be determined based on the thickness of the sections, the type of tissue, and its age.

2. The function of phosphomolybdic acid is twofold: on one hand, it differentiates collagen fibers that have been stained red into colorless or pale red, while leaving muscle fibers and cellulose still appearing bright red; on the other hand, it acts as a mordant for collagen fibers, facilitating their binding with Aniline Blue Stain Solution.

3. Treatment with Weak Acid Solution after Aniline Blue staining aims to remove the blue color from the cytoplasm, resulting in a vivid and clear staining. For tissues fixed with Zenker's solution, the treatment with Weak Acid Solution can be extended to 5 minutes.

4. Weak Acid Solution can enhance color clarity and vividness. If a large quantity is required, a 0.1-0.3% acetic acid solution can be prepared as a substitute.

Disclaimer

The reagent is only used in the field of scientific research, not suitable for clinical diagnosis or other purposes.