

Delafield Hematoxylin Stain Solution

Introduction

Delafield hematoxylin is identical to Ehrlich hematoxylin in that both are forms of naturally oxidized hematoxylin. This means that hematoxylin is exposed to light and air to undergo natural oxidation into hematein. This process is relatively slow, typically taking 3 to 6 months; however, the staining efficacy can be maintained for a long time.

The Delafield hematoxylin staining solution provides clear and detailed visualization of nuclear chromatin. The staining duration is relatively long, approximately 15-20 min. This staining solution is suitable for paraffin section staining in teaching and research settings but is not applicable to frozen section staining.

Storage

Store at room temperature protected from light, stable for 2 years.

Protocol

1. Materials Required but Not Included

- 1) Hydrochloric acid ethanol differentiation solution
- 2) Bluing solution (such as dilute ammonia water, lithium carbonate solution, etc.)
- 3) Series of ethanol solutions
- 4) Eosin staining solution
- 5) 4% paraformaldehyde

2. Paraffin Section Staining

- 1) Slice Dewaxing to Water
 - a) Xylene treatment twice, 5-10 min each time.
 - b) (Optional) Anhydrous ethanol treatment twice, 3-5 min each time.
 - c) 95% ethanol for 3-5 min, 90% ethanol for 3-5 min, 80% ethanol for 3-5 min.
 - d) Tap water or distilled water rinse for 1-3 min.
- 2) Staining

- a) Stain with Delafield Hematoxylin Stain Solution for 15-20 min.
- b) Rinse with tap water or distilled water for 5-10 sec.
- c) (Optional) Differentiate with hydrochloric acid ethanol for 2-5 sec, rinse with tap water for 20-30 sec.
- d) (Optional) Return to blue with bluing solution for 20-40 sec, rinse with tap water for 30-60 sec.
- e) Stain with eosin staining solution for 10-20 sec, rinse with distilled water for 1-5 sec.

3) Dehydration, Clearing, and Mounting

- a) 80% ethanol for 10-20 sec, 90% ethanol for 10-20 sec, 95% ethanol treatment twice, 1-2 min each time.
- b) Anhydrous ethanol treatment twice, 2-3 min each time.
- c) Xylene clearing three times, 2-3 min each time.
- d) Mount with neutral balsam.

3. Cell Staining

- 1) Fix with 4% paraformaldehyde for 10-20 min.
- 2) Rinse with tap water twice, 2 min each time.
- 3) Rinse with distilled water twice, 2 min each time.
- 4) The steps for staining, dewaxing, clearing, and mounting are the same as those for paraffin sections, but the action time should be shortened accordingly.

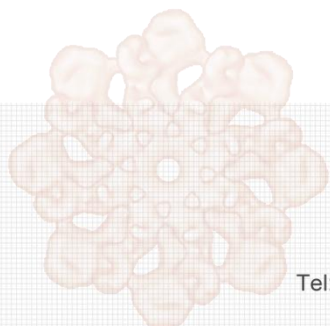
4. Staining Result

Nucleus	Blue
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Note

1. Slices should be thoroughly dewaxed. The series of ethanol solutions should be frequently replaced with fresh ones.
2. The differentiation time in hydrochloric acid ethanol should be determined based on the thickness of the slice, tissue type, and whether the slide is new or old. Additionally, the rinsing time with tap water after differentiation should be sufficient to completely remove the acid.
3. Bluing solutions commonly used include 0.2-1% ammonia water, Scott's bluing solution, or 0.1-1% lithium carbonate solution.
4. For your safety and health, please wear lab coats and gloves during the experiment.

5. For research use only. Not to be used in clinical diagnostic or clinical trials.



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