Hydrochloric Acid-Ethanol Fast Differentiation Solution (20×)

A743393

Storage: Room Temperature.

Introduction:

Differentiation refers to the process of removing excess stain bound to tissues using specific solutions after tissue staining. The solution used in this process is called a differentiation solution. In HE staining and other staining methods, 1% hydrochloric acid-ethanol is commonly used as a differentiation solution. Acids can disrupt the quinone structure of hematoxylin, separating tissues from the pigment and causing discoloration. Subsequent eosin staining ensures clear differentiation between cell nuclei and cytoplasm.

Hydrochloric acid-ethanol differentiation solution mainly consists of hydrochloric acid, ethanol, and deionized water. It is frequently used in hematoxylin staining, hematoxylin-eosin staining (HE staining), Masson staining, and other procedures involving hematoxylin. As a crucial auxiliary reagent, it can slowly reduce background hematoxylin staining in structures like cytoplasm while enhancing the clarity of hematoxylin staining in cell nuclei. Using this product ensures better hematoxylin staining results. Its basic principle is that under acidic conditions, it can effectively remove hematoxylin staining outside cell nuclei to reduce background. Meanwhile, it can appropriately lighten over-stained areas of cell nuclei, resulting in clearer staining.

Hydrochloric Acid-Ethanol Fast Differentiation Solution (20×) is a stock solution. It must be diluted with 70% ethanol before use. The diluted working solution requires a relatively short differentiation time, typically around 10 seconds. For even faster differentiation, you may consider purchasing Hydrochloric Acid-Ethanol Ultra-Fast Differentiation Solution or using 1% acidic ethanol differentiation solution. This reagent is for research use only and not intended for clinical diagnosis or other purposes.

Materials to Be Prepared by the User:

1. Tap water or distilled water, 4% paraformaldehyde or neutral formalin fixative, xylene or wax-immersion dewaxing and clearing solution.
2. A series of ethanol solutions, hematoxylin staining solution, eosin staining solution, bluing solution (e.g., dilute ammonia water, lithium carbonate solution), and neutral balsam.

Operating Procedures (HE Staining Steps, for Reference Only):

1. Preparation of Hydrochloric Acid-Ethanol Fast Differentiation Solution: Mix 5 parts of Hydrochloric Acid-Ethanol Fast Differentiation Solution (20×) with 95 parts of 70% ethanol.
2. Dewax the tissue to water.

3. Stain with hematoxylin staining solution for 3-8 minutes.
4. Rinse with tap water or distilled water for 5-10 seconds.
5. Differentiate with Hydrochloric Acid-Ethanol Fast Differentiation Solution for 5-15 seconds.
6. Rinse with tap water for 20-30 seconds.
7. Blue the tissue with bluing solution for 20-40 seconds or with warm water for 10 minutes.
8. Dehydrate with 80% ethanol for 30-60 seconds.
9. Stain with alcoholic eosin staining solution for 20-60 seconds.
10. Dehydrate with gradient ethanol solutions, clear with xylene or environment-friendly dewaxing and clearing solution, mount the slide with neutral balsam, and examine under a microscope.

Precautions:

1. The required differentiation time varies depending on different samples and staining requirements. For first-time use, it is recommended to test different differentiation times and determine the optimal duration based on microscopic observations.
2. Post-staining differentiation is an optional step, but it improves the clarity of nucleus-cytoplasm staining and enhances the visibility of nuclear membranes and chromatin morphology.
3. This product is volatile; store it in a sealed container. To avoid reduced effectiveness due to volatilization, use it as soon as possible after opening.
4. This product is corrosive. Operate with caution and take effective protective measures to avoid direct contact with the human body or corrosion of other items.
5. For your safety and health, wear a lab coat and disposable gloves during operation.
6. Use the reagent as soon as possible after opening to prevent adverse effects on subsequent experimental results.