Immunohistochemistry Enhancer

Cat #: B-IMWRS52

Size: 100 T

Storage: Store at 2°C~8°C protect from light

Intended use

This product is intended for Immunohistochemical staining, used in working with normal secondary antibodies.

Kit components:

Kit component	Volume	Storage
Enhancer	White dropper bottle, 10mL	2-8°C

Storage

Storage: Store sealed at 2°C to 8°C, for 1 year.

Suggested Protocol

1. Human tissue samples are first fixed with formaldehyde.(Fresh biopsy or surgical tissue samples fixed with formaldehyde for 8-24 hours).
2. The fixed tissues undergo a series of operations including dehydration to prepare wax blocks.
3. The tissue wax blocks are cut into section using a microtome.

4. Manual operation of immunohistochemistry experiment. The recommended routine steps are as follows:
   4.1. Take out the tissue section for the experiment and place them on glass slides. Put them in an oven at 60-65°C and bake for 1-1.5 hours.
   4.2. Place the section in the following staining dishes for gradient reactions:
   - Xylene 10 minutes, twice
   - Xylene 5 minutes
   - Absolute ethanol 5 minutes, twice
   - 95% ethanol 5 minutes
   - 85% ethanol 5 minutes
   - PBS wash for 3 minutes, three times
   4.3. Perform antigen retrieval (refer to the primary antibody supplier's instructions or use conventional methods such as high pressure, microwave etc.)
   4.4. Directly drop endogenous peroxidase blocker onto the tissue based on tissue size, typically 100 µL, until the tissue is completely covered. Incubate at room temperature for 10 minutes each, followed by PBS wash for 3 minutes, three times.
   This step is usually performed after mild antigen retrieval methods such as water bath or enzyme treatment. Alternatively, it can be added if the tissue itself contains a high level of endogenous peroxidase.
   4.5. Drop the primary antibody onto the tissue based on its size, typically 100 µL, until the tissue is completely covered. Incubate in a humid box for 1 hour. PBS wash for 3 minutes, three times. (The optimal reaction time for the primary antibody may vary slightly depending on the manufacturer.)
   4.6. Optional Step: Add 100 µL of **enhancer** onto the tissue, covering it completely, depending on the size of the tissue. Incubate at room temperature for 10 minutes, followed by PBS washing for 3 minutes × 3 times. This step is usually performed using an automated stainer. In manual procedures, the addition of enhancer can be optional based on the desired staining effect, as the enhancer can amplify the signal of the primary antibody from mouse origin by 1-10 times.
5. Add HRP secondary antibody conjugate, using a volume sufficient to cover all the tissue, typically 100 µL. Incubate at 37°C for 30 minutes.
6. DAB Staining: The DAB staining solution should be prepared and used immediately.
7. Terminate the color development by rinsing with tap water, followed by counterstaining with hematoxylin; differentiate and rinse in a blueing solution.
8. Dehydrate with alcohol, clear with xylene, mount with neutral resin, and examine the slides.

Precautions

1. This product is for research use only and should not be used for other purposes.
2. It should be used by professionals.
3. Apply appropriate protective measures to avoid contact of the reagents with the skin and eyes.
4. Dispose of waste liquids in an environmentally friendly manner and in accordance with relevant regulations.

Note:

The product listed herein is for research use only and is not intended for use in human or clinical diagnosis.