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NBP2-75981 Protocol

Immunohistochemistry Protocol for CD57 Antibody (NBP2-75981)

Immunohistochemistry Protocol

Specimen Collection and Preparation for Analysis

Each tissue section should be fixed with 10% neutral buffered formalin, cut to the applicable thickness (4um), and placed on a glass slide that is positively charged. The prepared slide may then be baked for a minimum of 30 minutes in a 53-65 degrees C oven (do not exceed 24 hours).

Recommended Staining Protocols

Manual Use:
1. Pretreatment: Perform heat-induced epitope retrieval (HIER) at pH 9 for 10 to 30 minutes.
2. Peroxide Block: Block in peroxidase blocking solution for 5 minutes at room temperature. (Not required if using Alkaline Phosphatase System.)
3. Primary Antibody: Apply antibody directly (Predilute) or dilute antibody at 1:100-1:200 (Concentrate) before applying. Incubate antibody for 10 to 30 minutes at room temperature.
4. Secondary Antibody: Incubate for 20 to 30 minutes at room temperature.
5. Substrate Development: Incubate DAB or Fast Red for 5 to 10 minutes at room temperature.
6. Counterstain: Counterstain with hematoxylin for 0.5 to 5 minutes, depending on the hematoxylin used. Rinse with distilled water and blueing solution for 30 seconds.
7. Dehydrate and apply coverslip.

Automated Staining System:
The stated primary antibody has been optimized and validated using the BOND-MAX fully automated IHC & ISH stainer manufactured by Leica Biosystems, applying IHC Protocol F. The following edits are recommended for the protocol:
a) Marker Incubation Time: 30 minutes
b) Heat-induced epitope retrieval (HIER) is recommended using Leica Bond ER Solution 2 for 30 minutes.
c) Move Peroxide Block step to after Polymer and before Mixed DAB Refine. For all other automated IHC staining systems, refer to the corresponding user manual for specific instructions.