

Immunohistochemistry Protocol

Product Information

Product Name: anti-PD-1 (human), mAb (AG-IHC001)

Catalog Number: AG-20B-6020

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: Dilute antibody as indicated in product datasheet before applying. Incubate antibody

for 10 to 30 minutes at room temperature.

4. Secondary Antibody: Incubate for 20 to 30 minutes at room temperature.

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.

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WARNING: Intended for research use only. This product is not intended or approved for human, diagnostics, therapeutic or veterinary use. Use of this product for human or animal testing is extremely hazardous and may result in disease, severe injury, or death. **MATERIAL SAFETY DATA:** Review the complete Material Safety Data Sheet before use.

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