

DESCRIPTION

Species Reactivity	Human
Specificity	Detects a synthetic peptide specific for human PD-1 around amino acid 40 in Direct ELISA.
Source	Monoclonal Mouse IgG Clone # 1109446
Purification	Protein A or G purified from cell culture supernatant
Immunogen	Synthetic Peptide Accession # Q15116
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose.

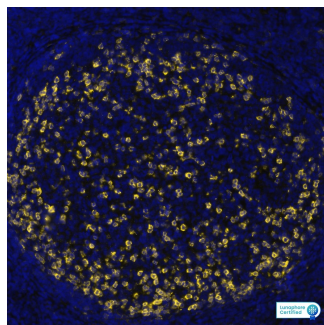
APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. [General Protocols](#) are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Western Blot	2 µg/mL	Human tonsil
Multiplex Immunofluorescence	20 µg/mL	Immersion fixed paraffin-embedded sections of human tonsil
Immunohistochemistry	3-25 µg/mL	Immersion fixed paraffin-embedded sections of human tonsil and lymph node

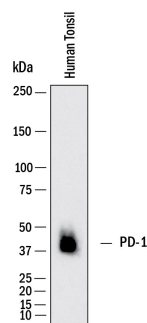
DATA

Multiplex Immunofluorescence



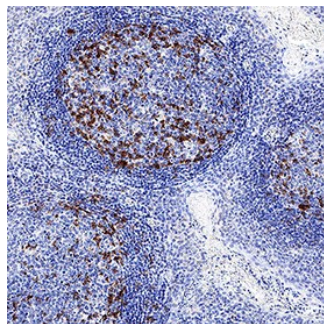
Detection of PD-1 in Human Tonsil via seqIF™ staining on COMET™ PD-1 was detected in immersion fixed paraffin-embedded sections of Human tonsil using Mouse Anti-Human PD-1, Monoclonal Antibody (Catalog #MAB11741) at 20µg/mL at 37 ° Celsius for 8 minutes. Before incubation with the primary antibody, tissue underwent an all-in-one dewaxing and antigen retrieval preprocessing using PreTreatment Module (PT Module) and Dewax and HIER Buffer H (pH 9; Epredia Catalog # TA-999-DHBH). Tissue was stained using the Alexa Fluor™ 647 Goat anti-Mouse IgG Secondary Antibody at 1:200 at 37 ° Celsius for 2 minutes. (Yellow; Lunaphore Catalog # [DR647MS](#)) and counterstained with DAPI (blue; Lunaphore Catalog # [DR100](#)). Specific staining was localized to the membrane. Protocol available in [COMET™ Panel Builder](#).

Western Blot



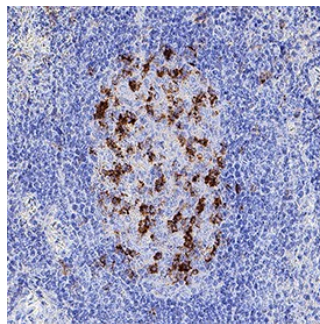
Detection of Human PD-1 by Western Blot. Western Blot shows lysates of human tonsil. PVDF membrane was probed with 2 µg/ml of Mouse Anti-Human PD-1 Monoclonal Antibody (Catalog # MAB11741) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # [HAF018](#)). A specific band was detected for PD-1 at approximately 40 kDa (as indicated). This experiment was conducted under reducing conditions and using [Western Blot Buffer Group 1](#).

Immunohistochemistry



Detection of PD-1 in Human Tonsil. PD-1 was detected in immersion fixed paraffin-embedded sections of human tonsil using Mouse Anti-Human PD-1 Monoclonal Antibody (Catalog # MAB11741) at 5 µg/ml for 1 hour at room temperature followed by incubation with the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # [VC001](#)) or the HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # [HAF007](#)). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # [VCTS021](#)). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to the membrane. View our protocol for [IHC Staining with VisUCyte HRP Polymer Detection Reagents](#).

Immunohistochemistry



Detection of PD-1 in Human Lymph Node. PD-1 was detected in immersion fixed paraffin-embedded sections of human lymph node using Mouse Anti-Human PD-1 Monoclonal Antibody (Catalog # MAB11741) at 5 µg/ml for 1 hour at room temperature followed by incubation with the Anti-Mouse IgG VisUCyte™ HRP Polymer Antibody (Catalog # [VC001](#)) or the HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # [HAF007](#)). Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # [VCTS021](#)). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to the membrane. View our protocol for [IHC Staining with VisUCyte HRP Polymer Detection Reagents](#).

PREPARATION AND STORAGE

Reconstitution	Reconstitute lyophilized material at 0.2 mg/ml in sterile PBS. For liquid material, refer to CoA for concentration.
Shipping	Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Programmed Death-1 receptor (PD-1), also known as CD279, is type I transmembrane protein belonging to the CD28 family of immune regulatory receptors (1). Other members of this family include CD28, CTLA-4, ICOS, and BTLA (2-5). Mature human PD-1 consists of a 148 amino acid (aa) extracellular region (ECD) with one immunoglobulin-like V-type domain, a 24 aa transmembrane domain, and a 95 aa cytoplasmic region. The human PD-1 ECD shares 65% aa sequence identity with the mouse PD-1 ECD. The cytoplasmic tail contains two tyrosine residues that form the immunoreceptor tyrosine-based inhibitory motif (ITIM) and immunoreceptor tyrosine-based switch motif (ITSM) that are important for mediating PD-1 signaling. PD-1 acts as a monomeric receptor and interacts in a 1:1 stoichiometric ratio with its ligands PD-L1 (B7-H1) and PD-L2 (B7-DC) (6, 7). PD-1 is expressed on activated T cells, B cells, monocytes, and dendritic cells while PD-L1 expression is constitutive on the same cells and also on nonhematopoietic cells such as lung endothelial cells and hepatocytes (8, 9). Ligand of PD-L1 with PD-1 induces co-inhibitory signals on T cells promoting their apoptosis, anergy, and functional exhaustion (10). Thus, the PD-1: PD-L1 interaction is a key regulator of the threshold of immune response and peripheral immune tolerance (11). Finally, blockade of the PD-1: PD-L1 interaction by either antibodies or genetic manipulation accelerates tumor eradication and shows potential for improving cancer immunotherapy (12, 13, 14).

References:

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