

Anti-PD-L1 Antibody [PSH08-27]

HA722971



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Dog
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 33 kDa
Clone number:	PSH08-27

Description:	Programmed death-ligand 1 (PD-L1) also known as cluster of differentiation 274 (CD274) or B7 homolog 1 (B7-H1) is a protein that in humans is encoded by the CD274 gene. Programmed death-ligand 1 (PD-L1) is a 40kDa type 1 transmembrane protein that has been speculated to play a major role in suppressing the adaptive arm of immune systems during particular events such as pregnancy, tissue allografts, autoimmune disease and other disease states such as hepatitis. Normally the adaptive immune system reacts to antigens that are associated with immune system activation by exogenous or endogenous danger signals. In turn, clonal expansion of antigen-specific CD8+ T cells and/or CD4+ helper cells is propagated. The binding of PD-L1 to the inhibitory checkpoint molecule PD-1 transmits an inhibitory signal based on interaction with phosphatases (SHP-1 or SHP-2) via Immunoreceptor Tyrosine-Based Switch Motif (ITSM). This reduces the proliferation of antigen-specific T-cells in lymph nodes, while simultaneously reducing apoptosis in regulatory T cells (anti-inflammatory, suppressive T cells) – further mediated by a lower regulation of the gene Bcl-2. PD-L1 is expressed on both hematopoietic and nonhematopoietic cells in tissues. However, the exact roles of PD-L1 on hematopoietic versus nonhematopoietic cells in modulating immune responses are unclear.
Immunogen:	Recombinant protein within dog PD-L1 aa 1-238.
Positive control:	BCMT transfected with PD-L1 cell lysate, BCMT cells transfected with PD-L1.
Subcellular location:	Membrane.
Database links:	SwissProt: E2RKZ5 Dog
Recommended Dilutions:	
WB	1:2,000
IF-Cell	1:2,500
IHC-P	1:5,000
Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.
Purity:	Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders: 0086-571-88062880

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Applications: WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

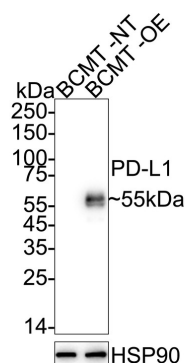


Fig1: Western blot analysis of PD-L1 on different lysates with Rabbit anti-PD-L1 antibody (HA722971) at 1/2,000 dilution.

Lane 1: BCMT cell lysate

Lane 2: BCMT transfected with PD-L1 cell lysate

Predicted band size: 33 kDa

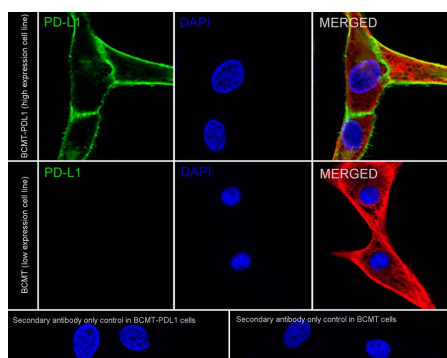
Observed band size: 55 kDa

Exposure time: 10 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA722971) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: Immunocytochemistry analysis of BCMT cells transfected with PD-L1 labeling PD-L1 with Rabbit anti-PD-L1 antibody (HA722971) at 1/2,500 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-PD-L1 antibody (HA722971) at 1/2,500 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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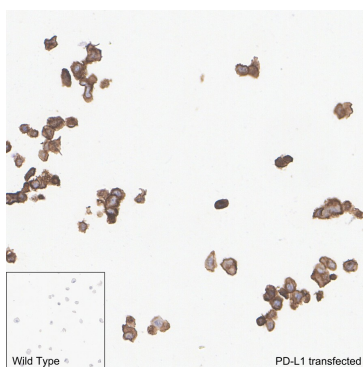


Fig3: Immunohistochemical analysis of paraffin-embedded BCMT cells transfected with PD-L1 with Rabbit anti-PD-L1 antibody (HA722971) at 1/5,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA722971) at 1/5,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Zhang YC et al. What role does PDL1 play in EMT changes in tumors and fibrosis? *Front Immunol.* 2023 Aug
2. Tran-Nguyen VK et al. Structure-based virtual screening for PDL1 dimerizers: Evaluating generic scoring functions. *Curr Res Struct Biol.* 2022 Jun

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