# Anti-PD-L1 Antibody [PSH13-23] - BSA and Azide free HA751459

| Product Type:<br>Species reactivity:<br>Applications:<br>Molecular Wt:<br>Clone number: | Recombinant Rabbit multiclonal IgG, primary antibodies<br>Human, Mouse<br>WB<br>Predicted band size: 33 kDa<br>PSH13-23                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              |
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| Description:                                                                            | PD-L1 (programmed-death ligand 1; CD274), is a transmembrane protein constitutionally expressed on a variety of cell types, including antigen presenting cells (dendritic cells and histiocytes) and some non-lymphoid tissues (heart and lung). Binding of PD-L1 to PD-1 (programmed-death 1; CD279) expressed by activated T-cells, inhibits their function, causing negative feedback control of immunological reactions, thus impeding inflammation and autoimmunity. Tumour cells may express PD-L1, which binds to PD-1 allowing cancer cells to evade the attack of T-cells. Blockade of the PD-1/PD-L1 pathway has now shown useful in therapy of multiple cancer types, causing durable tumour regressions in a substantial proportion of otherwise treatment refractory cases of melanoma, and carcinomas of e.g., lung, kidney, and urinary tract. Patients without tumour PD-L1 expression can also derive benefit from blocking agents (studies across multiple cancer types demonstrate a pooled response rate of 48% in patients with PD-L1-positive tumours compared to 15% in PD-L1-negative tumours). Tonsil and placenta can be used as positive and negative tissue controls. However, tonsil is found to be superior to placenta, as tonsil displayes a range of PD-L1 expression levels. Tonsil displayes the following reaction pattern: No staining reaction in the vast majority of lymphocytes including mantle zone and germinal centre B-cells, no staining reaction in superficial epithelial cells, a weak to moderate, typically punctuated membranous staining reaction of the majority of germinal centre macrophages and finally a moderate to strong staining reaction of the majority of epithelial crypt cells. |
| Immunogen:                                                                              | Synthetic peptide within human PD-L1 aa 260-290 (Cytoplasmic).                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                       |
| Positive control:                                                                       | MDA-MB-231 cell lysate, U-87 MG cell lysate, RAW264.7 cell lysate, RAW264.7 treated with 10 $\mu$ g/mL LPS for 8 hours cell lysate, J774A.1 cell lysate, J774A.1 treated with 1 $\mu$ g/mL LPS for 24 hours cell lysate, Mouse spleen tissue lysate, Mouse lung tissue lysate.                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                       |
| Subcellular location:                                                                   | Cell membrane, Early endosome membrane, Recycling endosome membrane, Nucleus.                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                        |
| Database links:                                                                         | SwissProt: Q9NZQ7 Human   Q9EP73 Mouse                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                               |
| Recommended Dilutions:<br>WB                                                            | 1:2,000                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              |
| Storage Buffer:                                                                         | PBS (pH7.4).                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |
| Storage Instruction:                                                                    | Store at +4 $^\circ\!\!\!C$ after thawing. Aliquot store at -20 $^\circ\!\!\!C$ . Avoid repeated freeze / thaw cycles.                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                               |
| Purity:                                                                                 | Protein A affinity purified.                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                         |

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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





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Fig1: Western blot analysis of PD-L1 on different lysates with Rabbit anti-PD-L1 antibody (HA751459) at 1/2,000 dilution.

Lane 1: MDA-MB-231 cell lysate Lane 2: MCF7 cell lysate (negative) Lane 3: U-87 MG cell lysate Lane 4: HepG2 cell lysate (negative)

Lysates/proteins at 20 µg/Lane.

Predicted band size: 33 kDa Observed band size: 45-55 kDa

Exposure time: 20 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 (HA751459) 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: Western blot analysis of PD-L1 on different lysates with Rabbit anti-PD-L1 antibody (HA751459) at 1/2,000 dilution.

Lane 1: RAW264.7 cell lysate

Lane 2: RAW264.7 treated with 10µg/mL LPS for 8 hours cell lysate

Lane 3: J774A.1 cell lysate

Lane 4: J774A.1 treated with 1µg/mL LPS for 24 hours cell lysate Lane 5: Mouse spleen tissue lysate

Lane 6: Mouse lung tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 33 kDa Observed band size: 45-55 kDa

Exposure time: 59 seconds; ECL: K1802; 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 (HA751459) 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.

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

#### **Background References**

- 1. Lei Q et al. Resistance Mechanisms of Anti-PD1/PDL1 Therapy in Solid Tumors. Front Cell Dev Biol. 2020 Jul
- Tamene W et al. PDL1 expression on monocytes is associated with plasma cytokines in Tuberculosis and HIV. PLoS One. 2021 Oct

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