Mouse PD-L1/B7-H1 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF1019

DESCRIPTION

Species Reactivity	Mouse
Specificity	Detects mouse PD-L1/B7-H1 in direct ELISAs and Western blots. In direct ELISAs, approximately 20% cross-reactivity with recombinant human PD-L1/B7-H1 is observed.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant mouse PD-L1/B7-H1 Phe19-Thr238 Accession # Q9EP73
Formulation	Lyophilized from a 0.2 μm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 μm filtered solution in PBS.

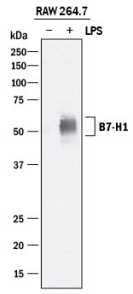
APPLICATIONS

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	0.5 µg/mL	See Below
Flow Cytometry	0.25 µg/10^6 cells	See Below
Immunohistochemistry	5-15 µg/mL	Perfusion fixed frozen sections of mouse small intestine (Peyer's patch) and thymus
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

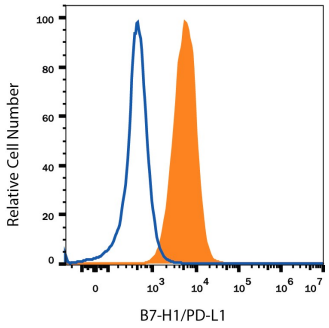
DATA

Western Blot



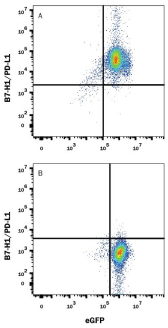
Detection of Mouse PD-L1/B7-H1 by Western Blot. Western blot shows lysates of RAW 264.7 mouse monocyte/macrophage cell line untreated (-) or treated (+) with 10 µg/mL LPS for 4 hours. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Mouse PD-L1/B7-H1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1019) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for PD-L1/B7-H1 at approximately 50-55 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Flow Cytometry



Detection of PD-L1/B7-H1 in RAW 264.7 Mouse Cell Line by Flow Cytometry. RAW 264.7 mouse monocyte/macrophage cell line either treated with LPS overnight (filled histogram) or untreated (open histogram) was stained with Goat Anti-Mouse PD-L1/B7-H1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1019), followed by Allophycocyanin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0108). View our protocol for Staining Membrane-associated Proteins.

Flow Cytometry



Detection of PD-L1/B7-H1 in HEK293 Human Cell Line Transfected with Mouse PD-L1/B7-H1 and eGFP by Flow Cytometry. HEK293 human embryonic kidney cell line transfected with either (A) mouse PD-L1/B7-H1 or (B) irrelevant transfectants and eGFP was stained with Goat Anti-Mouse PD-L1/B7-H1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF1019) followed by Allophycocyanin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0108). Quadrant markers were set based on control antibody staining (Catalog # AB-108-C). View our protocol for Staining Membrane-associated Proteins.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
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.

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Global bio-techne.com info@bio-techne.com techsupport@bio-techne.com TEL +1 612 379 2956
USA TEL 800 343 7475 Canada TEL 855 668 8722 China TEL +86 (21) 52380373
Europe | Middle East | Africa TEL +44 (0)1235 529449

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BACKGROUND

Mouse B7 homolog 1(B7-H1), also called programmed death ligand 1 (PD-L1) and programmed cell death 1 ligand 1 (PDCD1L1), is a member of the B7 family of proteins that provide signals for regulating T-cell activation and tolerance (1-4). Other family members include B7-1, B7-2, B7-H2, B7-H3 and PD-L2. B7 proteins are immunoglobulin (Ig) superfamily members with extracellular Ig-V-like and Ig-C-like domains and a short cytoplasmic region. Among the family members, they share from 20-40% amino acid (aa) sequence identity. The cloned mouse B7-H1/PD-L1 cDNA encodes a 290 aa type I membrane precursor protein with a putative 18 aa signal peptide, a 220 aa extracellular region containing one V-like and one C-like Ig domain, a 22 aa transmembrane region, and a 30 aa cytoplasmic domain. Mouse and human B7-H1/PD-L1 share approximately 70% aa sequence identity. B7-H1/PD-L1 is one of two ligands for programmed death-1 (PD-1), a member of the CD28 family of immunoreceptors. The other identified ligand is PD-L2. Mouse B7-H1/PD-L1 and PD-L2 share approximately 34% aa sequence identity and have similar functions. B7-H1/PD-L1 is constitutively expressed in various lymphoid and non-lymphoid organs including placenta, heart, pancreas, lung, liver, and endothelium (1-4). The expression of B7-H1/PD-L1 is detected on B cells, T cells, monocytes, dendritic cells and thymic epithelial cells. IFN-γ treatment induces B7-H1/PD-L1 expression in monocytes, dendritic cells, and endothelial cells. B7-H1/PD-L1 expression is also upregulated in a variety of tumor cell lines. On previously activated T cells, B7-H1/PD-L1 interaction with PD-1 inhibits TCR-mediated proliferation and cytokine production, suggesting an inhibitory role in regulating immune responses. In contrast, a costimulatory function for the PD-1 ligands on resting T cells has also been reported (1-4).

References:

1. Tamura, H. *et al.* (2001) Blood **97**:1809.
2. Freeman, G. *et al.* (2000) J. Exp. Med. **192**:1027.
3. Sharpe, A.H. and G. J. Freeman (2002) Nat. Rev. Immunol. **2**:116.
4. Coyle, A. and J. Gutierrez-Ramos (2001) Nat. Immunol. **2**:203.

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Europe | Middle East | Africa TEL +44 (0)1235 529449