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EB06621 - Goat Anti-CD274 / PD-L1 Antibody

Size: 100µg specific antibody in 200µl

Target Protein

Principal Names: CD274 antigen, PD-L1, PDCD1LG1, B7-H, B7H1, PDL1, PDCD1L1, programmed cell death 1 ligand 1, PDL1, HGNC:17635, CD274, CD274 molecule, MGC142294, MGC142296
Official Symbol: CD274
Accession Number(s): NP_054862.1; NP_001254635.1
Human GeneID(s): 29126
Important Comments: This antibody is expected to recognize reported isoforms a and b (NP_054862.1; NP_001254635.1) only.

Immunogen

Peptide with sequence CKKQSDTHLEET, from the C Terminus of the protein sequence according to NP_054862.1; NP_001254635.1.

Please note the peptide is available for sale.

Purification and Storage

Purified from goat serum by ammonium sulphate precipitation followed by antigen affinity chromatography using the immunizing peptide.
Supplied at 0.5 mg/ml in Tris saline, 0.02% sodium azide, pH7.3 with 0.5% bovine serum albumin.
Aliquot and store at -20°C. Minimize freezing and thawing.

Applications Tested

Peptide ELISA: antibody detection limit dilution 1:128000.
Western blot: Approx 40kDa band observed in Human Heart lysates. The observed molecular weight corresponds to glycosylation (calculated MW of 33.3kDa according to NP_054862.1).. An additional band was also consistently observed at 55kDa, and was successfully blocked by incubation with the immunizing peptide. Preliminary testing also showed the 37+55kDa bands in lysates of cell lines, A549, Daudi, HeLa, HepG2 and Jurkat Recommended concentration: 0.03-0.1µg/ml. Primary incubation 1 hour at room temperature.
IHC: In paraffin embedded Human Placenta shows membranous staining of cytotrophoblasts. Recommended concentration: 2-4µg/ml.

Immunofluorescence: Strong expression of the protein seen in the cytoplasm of A431 and U2OS cells. Recommended concentration: 10µg/ml.

Flow Cytometry: Flow cytometric analysis of Jurkat cells. Recommended concentration: 10ug/ml.

Species Reactivity

Tested: Human
Expected from sequence similarity: Human

Specific References

This antibody has been successfully used in Western blot in Human:
Guozhi Xia, Xiaopu Zheng, Xinye Yao, Xiaowei Yao, Zhongwei Liu, Junkui Wang. Expression of programmed cell death-1 and its ligand B7 homolog 1 in peripheral blood lymphocytes from patients with peripartum cardiomyopathy.
Clin Cardiol. 2016 Dec 27.

PMID: 28026044

This antibody has been successfully used in Western blot and IHC in Human:
Chen J, Li G, Meng H, Fan Y, Song Y, Wang S, Zhu F, Guo C, Zhang L, Shi Y.
Upregulation of B7-H1 expression is associated with macrophage infiltration in hepatocellular carcinomas.
Cancer Immunol Immunother. 2012 Jan;61(1):101-8.
PMID: 21853301

EB06621 Flow cytometric analysis of paraformaldehyde fixed Jurkat cells (blue line), permeabilized with 0.5% Triton. Primary incubation 1hr (10ug/ml) followed by Alexa Fluor 488 secondary antibody (1ug/ml). IgG control: Unimmunized goat IgG (black line) followed by Alexa Fluor 488 secondary antibody.

EB06621 (2µg/ml) staining of paraffin embedded Human Placenta. Microwaved antigen retrieval with citrate buffer pH 6, HRP-staining.

EB06621 Negative Control showing staining of paraffin embedded Human Placenta, with no primary antibody.

EB06621 Immunofluorescence analysis of paraformaldehyde fixed A431 cells, permeabilized with 0.15% Triton. Primary incubation 1hr (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml), showing cytoplasmic staining. The nuclear stain is DAPI (blue). Negative control: Unimmunized goat IgG (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml).

EB06621 Immunofluorescence analysis of paraformaldehyde fixed U2OS cells, permeabilized with 0.15% Triton. Primary incubation 1hr (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml), showing cytoplasmic staining. The nuclear stain is DAPI (blue). Negative control: Unimmunized goat IgG (10ug/ml) followed by Alexa Fluor 488 secondary antibody (2ug/ml).

EB06621 (0.3µg/ml) staining of Human Heart (A) lysate + Blocking peptide (B) (35µg protein in RIPA buffer). Detected by chemiluminescence.