abcam

Product datasheet

Anti-PD-L1 antibody [EPR20529] ab213480

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-PD-L1 antibody [EPR20529]
Description	Rabbit monoclonal [EPR20529] to PD-L1
Host species	Rabbit
Specificity	This antibody is not suitable in IHC-FR for mouse samples
Tested applications	**Suitable for:** ICC/IF, IP, WB **Unsuitable for:** IHC-Fr or IHC-P
Species reactivity	**Reacts with:** Mouse
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Mouse placenta lysate, HEK mPDL1 cell lysate, whole cell lysate from RAW 264.7 cells treated with Interferon-gamma and whole cell lysate from MEF cells treated with Interferon-gamma. IP: Mouse placenta lysate. ICC/IF: RAW 264.7 cells treated with IFN-? (100 ng/ml) for 24 hours.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified

Clonality	Monoclonal
Clone number	EPR20529
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab213480 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

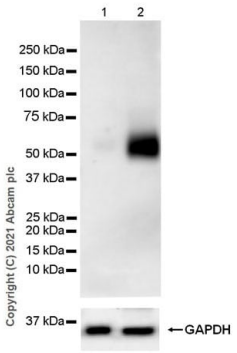
Application	Abreviews	Notes
ICC/IF		Use at an assay dependent concentration.
IP		1/30.
WB	★★★★★ (3)	1/1000. Detects a band of approximately 40-60 kDa (predicted molecular weight: 33 kDa).

Application notes Is unsuitable for IHC-Fr or IHC-P.

Target

Function	Involved in the costimulatory signal, essential for T-cell proliferation and production of IL10 and IFNG, in an IL2-dependent and a PDCD1-independent manner. Interaction with PDCD1 inhibits T-cell proliferation and cytokine production.
Tissue specificity	Highly expressed in the heart, skeletal muscle, placenta and lung. Weakly expressed in the thymus, spleen, kidney and liver. Expressed on activated T- and B-cells, dendritic cells, keratinocytes and monocytes.
Sequence similarities	Belongs to the immunoglobulin superfamily. BTN/MOG family. Contains 1 Ig-like C2-type (immunoglobulin-like) domain. Contains 1 Ig-like V-type (immunoglobulin-like) domain.
Cellular localization	Cell membrane and Endomembrane system.

Images



Western blot - Anti-PD-L1 antibody [EPR20529] (ab213480)

All lanes : Anti-PD-L1 antibody [EPR20529] (ab213480) at 1/5000 dilution

Lane 1 : Untreated RAW264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate
Lane 2 : RAW264.7 treated with 100 ng/ml IFN gamma for 48 h, whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

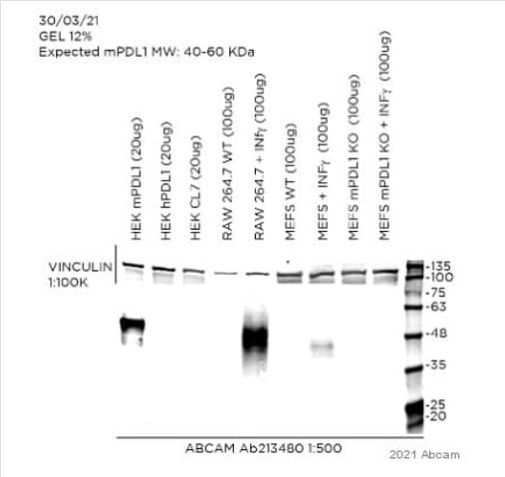
Predicted band size: 33 kDa
Observed band size: 40-60 kDa

Exposure time: 37 seconds

This WB was performed by using YCA-R20529(BF)-111A H3L2 YR120921PJ, 1:5000 dilution. Working concentration: 0.402 µg/ml.

Blocking buffer and concentration: 5% NFDM/TBST

Diluting buffer and concentration: 5% NFDM/TBST



Western blot - Anti-PD-L1 antibody [EPR20529] (ab213480)

This image is courtesy of an anonymous collaborator.

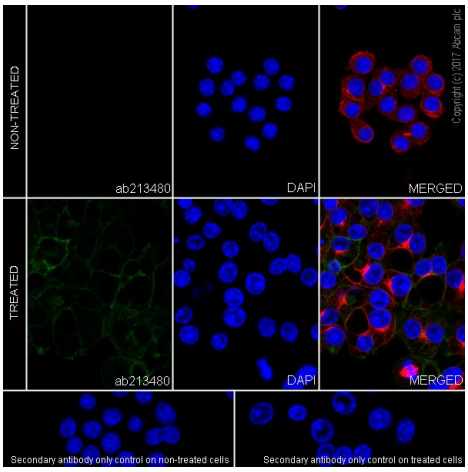
All lanes : Anti-PD-L1 antibody [EPR20529] (ab213480) at 100 µg

Lane 1 : HEK mPDL1 cell lysate at 20 µg
Lane 2 : HEK hPDL1 cell lysate at 20 µg
Lane 3 : HEK CL7 at 20 µg
Lane 4 : RAW 264.7 Wild-type (Mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate at 100 µg
Lane 5 : RAW 264.7 (Mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate treated with Interferon-gamma at 100 µg
Lane 6 : MEF Wild-type (Mouse embryonic fibroblast cell line) whole cell lysate at 100 µg
Lane 7 : MEF (Mouse embryonic fibroblast cell line) whole cell lysate treated with Interferon-gamma at 100 µg
Lane 8 : MEF mPDL1 KO cell lysate at 100 µg
Lane 9 : MEF mPDL1 KO treated with Interferon-gamma at 100 µg

Predicted band size: 33 kDa
Observed band size: 48 kDa

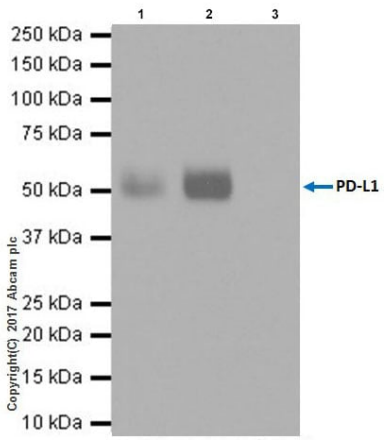
Exposure time: 5 minutes

The mAb is specific for mouse mPDL1 and does not recognise hPDL1. As expected a specific 48kDa band is observed in RAW264.7 and MEFS cell extract after Interferon-gamma treatment. No bands are observed in RAW264.7 and MEFS WT and in MEFS KO with and without Interferon-gamma treatment.



Immunocytochemistry/ Immunofluorescence - Anti-PD-L1 antibody [EPR20529] (ab213480)

Ab213480 staining PD-L1 in RAW264.7 from Mouse macrophage cell line transformed with Abelson murine leukemia virus by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with 100% Methanol. Samples were incubated with ab213480 at 0.4 µg/ml. AlexaFluor®488 Goat anti-Rabbit (**ab150077**) was used as the secondary antibody at 2 µg/ml. Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)(**ab196889**) was used as counterstain at 2.5 µg/ml. DAPI used as nuclear counterstain. Confocal image showing membranous staining on RAW 264.7 cells treated with IFN-γ (100 ng/ml) for 24 hours.



Immunoprecipitation - Anti-PD-L1 antibody [EPR20529] (ab213480)

PD-L1 was immunoprecipitated from 0.35 mg of Mouse placenta lysate with ab213480 at 1/30 dilution.

Western blot was performed from the immunoprecipitate using ab213480 at 1/500 dilution.

VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1,000 dilution.

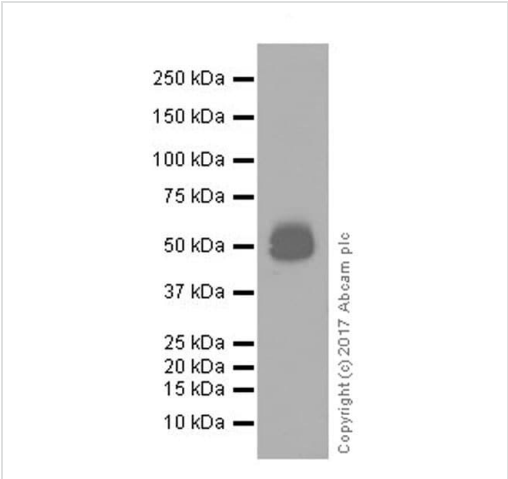
Lane 1: Mouse placenta lysate, 10 µg (Input).

Lane 2: ab213480 IP in mouse placenta lysate.

Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab213480 in mouse placenta lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3 seconds.



Western blot - Anti-PD-L1 antibody [EPR20529] (ab213480)

Anti-PD-L1 antibody [EPR20529] (ab213480) at 1/1000 dilution + Mouse placenta lysate at 10 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 33 kDa

Observed band size: 40-60 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been described in the literature. PMID: 15353579.

Anti-PD-L1 antibody [EPR20529] (ab213480)

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