abcam

Product datasheet

Anti-PD1 (phospho Y248) antibody [EPR19821] ab206378

Recombinant

RabMAb

4 Images

Overview

Product name Anti-PD1 (phospho Y248) antibody [EPR19821]

Description Rabbit monoclonal [EPR19821] to PD1 (phospho Y248)

Host species Rabbit

Tested applications Suitable for: WB, Dot blot, IP

Species reactivity Reacts with: Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: HEK-293T transfected with a PD1 (WT) plus a 25 kDa fusion protein expression vector, then

treated with 100 μ M pervanadate for 10 minutes whole cell lysate. IP: HEK-293T transfected with PD1 (WT) plus 25 kDa fusion protein expression vector, then treated with 100 μ M pervanadate for

10 minutes whole cell lysate.

General notesThis 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: PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EPR19821

1

Isotype IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab206378 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
WB		1/1000. Predicted molecular weight: 32 kDa.
Dot blot		1/1000.
IP		1/30.

Target

Function Possible cell death inducer, in association with other factors.

Involvement in disease Genetic variation in PDCD1 is associated with susceptibility to systemic lupus erythematosus

type 2 (SLEB2) [MIM:605218]. Systemic lupus erythematosus is a chronic, inflammatory and often febrile multisystemic disorder of connective tissue. It affects principally the skin, joints, kidneys and serosal membranes. It is thought to represent a failure of the regulatory mechanisms of the

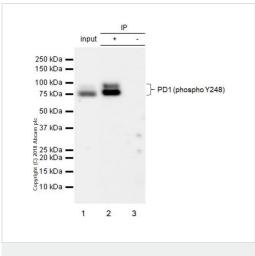
autoimmune system.

Sequence similaritiesContains 1 lg-like V-type (immunoglobulin-like) domain.

Developmental stage Induced at programmed cell death.

Cellular localization Membrane.

Images



Immunoprecipitation - Anti-PD1 (phospho Y248) antibody [EPR19821] (ab206378)

PD1 (phospho Y248) was immunoprecipitated from 0.35 mg of HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) transfected with a PD1 (WT) plus 25 kDa fusion protein expression vector, then treated with 100 μ M pervanadate for 10 minutes whole cell lysate, with ab206378 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab206378 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1/5000 dilution.

Lane 1: HEK-293T transfected with a PD1 (WT) plus 25 kDa fusion protein expression vector, then treated with 100 μ M pervanadate for 10 minutes whole cell lysate 10 μ g (Input).

Lane 2: ab206378 IP in HEK-293T transfected with a PD1 (WT) plus 25 kDa fusion protein expression vector, then treated with 100

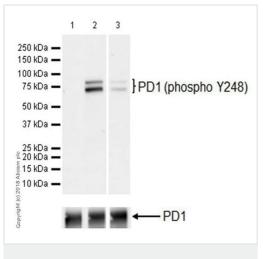
μM pervanadate for 10 minutes whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab206378 in HEK-293T transfected with a PD1 (WT) plus 25 kDa fusion protein expression vector, then treated with 100 μ M pervanadate for 10 minutes whole cell lysate.

Exposure time: 30 seconds

Blocking and diluting buffer and concentration: 5% NFDM/TBST

The observed bands around 75 kDa comprise PD-1 plus the 25 kDa fusion protein. The plasmids were kindly provided by our collaborator Dr. Liang Chen, NIBS.



Western blot - Anti-PD1 (phospho Y248) antibody [EPR19821] (ab206378)

All lanes : Anti-PD1 (phospho Y248) antibody [EPR19821] (ab206378) at 1/1000 dilution

Lane 1: Untreated-HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate transfected with a PD1 (WT) plus a 25 kDa fusion protein expression vector

Lane 2 : HEK-293T transfected with a PD1 (WT) plus a 25 kDa fusion protein expression vector, then treated with 100 μ M pervanadate for 10 minutes whole cell lysate

Lane 3: HEK-293T transfected with PD1 (WT) plus a 25 kDa fusion protein expression vector, then treated with 100 μ M pervanadate for 10 minutes, whole cell lysate (20 μ g). Following the transfer to PVDF, the membrane was treated with alkaline phosphatase for 1h at 37°C.

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 32 kDa

Observed band size: 70-75 kDa

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

The observed bands at around 75 kDa are PD-1 plus the 25 kDa

fusion protein. The expression profile observed is consistent with what has been described in the literature (PMID: 22641383). The plasmids were kindly provided by our collaborator Dr. Liang Chen NIBS.

Dot blot analysis of PD1 (phospho Y248) labeled with ab206378 at

1 2
5ng
1ng
0.1ng
0.01ng

1/1000 dilution.

Lane 1: PD1 (phospho Y248) peptide.

Lane 2: PD1 non-phospho peptide.

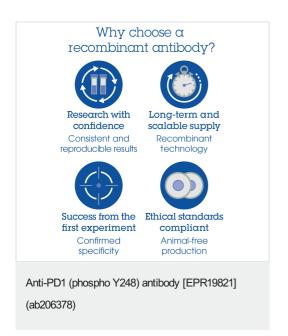
Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution was used as secondary antibody.

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 1 minute.

The blot was developed on a BIO-RAD[®] ChemiDoc™ MP instrument.

Dot Blot - Anti-PD1 (phospho Y248) antibody [EPR19821] (ab206378)



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