Anti-CD2 Antibody [PSH05-85] - BSA and Azide free

HA751033

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IHC-P, IF-Cell, FC, IF-Tissue
Molecular Wt:	Predicted band size: 39 kDa
Clone number:	PSH05-85

Description: CD2 is a transmembrane glycoprotein expressed early in thymocyte development and present on most circulating T cells. CD2 plays a role in T cell adhesion through binding to its ligand CD58 (LFA-3). Stimulation of CD2 also leads to T cell activation and proliferation. T cells from mice deficient in both CD2 and CD28 have severe defects in T cell activation and function, while T cells deficient in either CD2 or CD28 are still capable of mounting a response, suggesting that CD2 and CD28 may have overlapping functions and may be able to compensate for each other. In addition, engagement of CD2 and CD58 was recently demonstrated to be the primary costimulatory signal in T cells that lack CD28. CD2 expression also distinguishes a subset of plasmacytoid dendritic cells found in tumors and tonsils that express lysozyme, higher levels of IL-12 p40, and higher levels of CD80.

Immunogen: Recombinant protein within Human CD2 aa 25-209 (Extracellular).

Positive control: Jurkat cell lysate, human T cell lymphoma tissue, human appendix tissue, human spleen tissue, Jurkat.

Subcellular location: Cell membrane.

Database links: SwissProt: P06729 Human

Recommended Dilutions:

WB	1:1,000
IHC-P	1:100
IF-Cell	1:100
FC	1:1,000
IF-Tissue	1:50

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

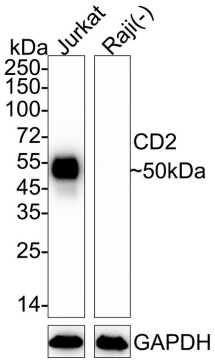

Fig1: Western blot analysis of CD2 on different lysates with Rabbit anti-CD2 antibody (HA751033) at 1/1,000 dilution.

Lane 1: Jurkat cell lysate
Lane 2: Raji cell lysate (negative)

Lysates/proteins at 20 µg/Lane.

Predicted band size: 39 kDa
Observed band size: 39-55 kDa

Exposure time: 25 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 (HA751033) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

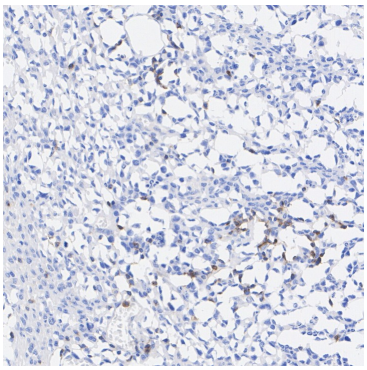

Fig2: Immunohistochemical analysis of paraffin-embedded human T cell lymphoma tissue with Rabbit anti-CD2 antibody (HA751033) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751033) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

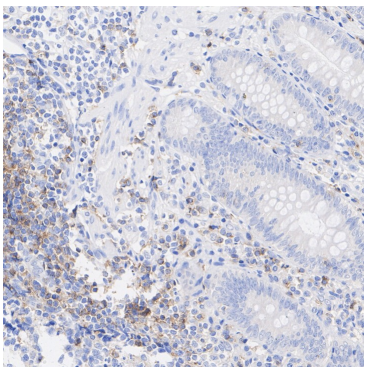

Fig3: Immunohistochemical analysis of paraffin-embedded human appendix tissue with Rabbit anti-CD2 antibody (HA751033) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751033) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

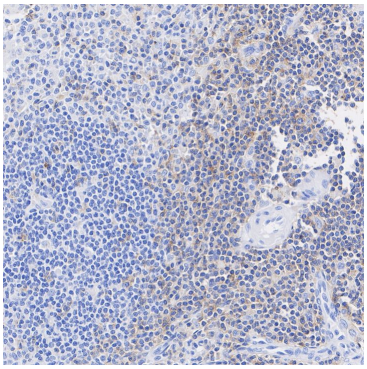

Fig4: Immunohistochemical analysis of paraffin-embedded human spleen tissue with Rabbit anti-CD2 antibody (HA751033) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751033) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

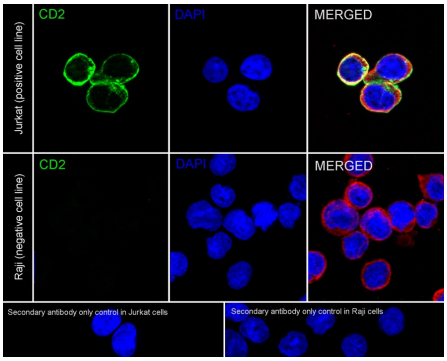

Fig5: Immunocytochemistry analysis of Jurkat (positive) and Raji (negative) labeling CD2 with Rabbit anti-CD2 antibody (HA751033) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-CD2 antibody (HA751033) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

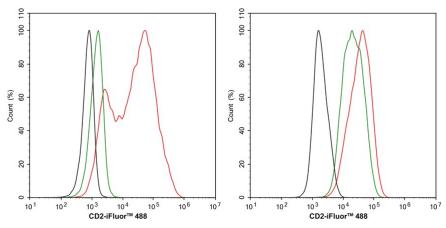

Fig6: Flow cytometric analysis of Jurkat (left, positive) and Raji (right, negative) cells labeling CD2.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA751033, 1μg/mL) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

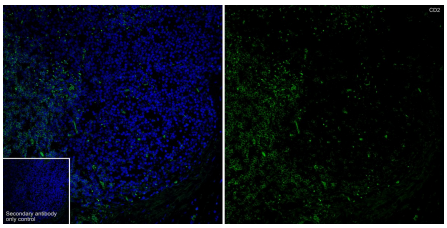

Fig7: Immunofluorescence analysis of paraffin-embedded human tonsil tissue labeling CD2 with Rabbit anti-CD2 antibody (HA751033) at 1/50 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA751033, green) at 1/50 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

Background References

1. Judith Leitner, Dietmar Herndler-Brandstetter, Gerhard J Zlabinger, Beatrix Grubeck-Loebenstein, Peter Steinberger. CD58/CD2 Is the Primary Costimulatory Pathway in Human CD28-CD8+ T Cells. J Immunol. 2015 Jul 15;195(2):477-87.
2. Toshimichi Matsui, John E Connolly, Mark Michnevitz, Damien Chaussabel, Chun-I Yu, Casey Glaser, Sasha Tindle, Marc Pypaert, Heidi Freitas, Bernard Piqueras, Jacques Banchereau, A Karolina Palucka. CD2 distinguishes two subsets of human plasmacytoid dendritic cells with distinct phenotype and functions. J Immunol. 2009 Jun 1;182(11):6815-23.