Anti-CD47 Antibody [PSH10-31] - BSA and Azide free

HA751348

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, FC
Molecular Wt:	Predicted band size: 35 kDa
Clone number:	PSH10-31

Description: CD47 (Cluster of Differentiation 47) also known as integrin associated protein (IAP) is a transmembrane protein that in humans is encoded by the CD47 gene. CD47 belongs to the immunoglobulin superfamily and partners with membrane integrins and also binds the ligands thrombospondin-1 (TSP-1) and signal-regulatory protein alpha (SIRPα). CD-47 acts as a don't eat me signal to macrophages of the immune system which has made it a potential therapeutic target in some cancers, and more recently, for the treatment of pulmonary fibrosis. CD47 is involved in a range of cellular processes, including apoptosis, proliferation, adhesion, and migration. Furthermore, it plays a role in insulin secretion, as well as immune and angiogenic responses. CD47 is ubiquitously expressed in human cells and has been found to be overexpressed in many different tumor cells. Expression in equine cutaneous tumors has been reported as well.

Immunogen: Recombinant protein within human CD47 aa 1-150.

Positive control: Jurkat cell lysate, HDLM-2 cell lysate, U-937 cell lysate, A549 cell lysate, NIH:OVCAR-3 cell lysate, MOLT-4 cell lysate, Jurkat.

Subcellular location: Cell membrane.

Database links: SwissProt: Q08722 Human

Recommended Dilutions:

WB	1:2,000
IF-Cell	1:250
FC	1:1,000

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

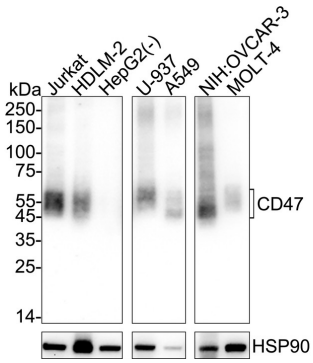

Fig1: Western blot analysis of CD47 on different lysates with Rabbit anti-CD47 antibody (HA751348) at 1/2,000 dilution.

Lane 1: Jurkat cell lysate
Lane 2: HDLM-2 cell lysate
Lane 3: HepG2 cell lysate (negative)
Lane 4: U-937 cell lysate
Lane 5: A549 cell lysate
Lane 6: NIH:OVCAR-3 cell lysate
Lane 7: MOLT-4 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 35 kDa
Observed band size: 45-55 kDa

Exposure time: Lane 1-5: 3 minutes; Lane 6-7: 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 (HA751348) at 1/2,000 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.

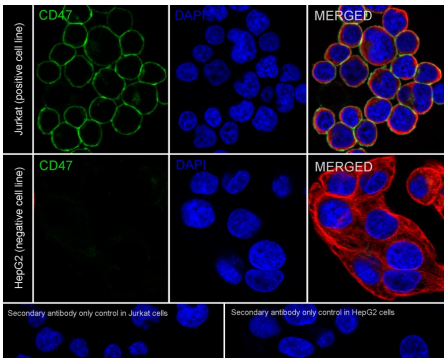

Fig2: Immunocytochemistry analysis of Jurkat (positive) and HepG2 (negative) labeling CD47 with Rabbit anti-CD47 antibody (HA751348) at 1/250 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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-CD47 antibody (HA751348) at 1/250 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 (HA601187, 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.

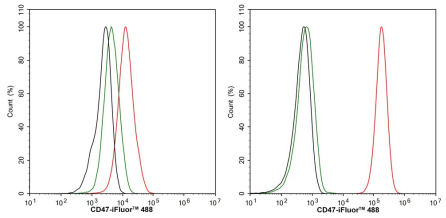

Fig3: Flow cytometric analysis of HepG2 (left, negative) and Jurkat (right, positive) cells labeling CD47.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA751348, 1/1,000) (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).

Background References

1. Logtenberg MEW et al. The CD47-SIRPalpha Immune Checkpoint. Immunity. 2020 May
2. van Duijn A et al. CD47/SIRPalpha axis: bridging innate and adaptive immunity. J Immunother Cancer. 2022 Jul