Anti-CD20 Antibody [PSH08-12] - BSA and Azide free

HA751207

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 33 kDa
Clone number:	PSH08-12

Description: B-lymphocyte antigen CD20 or CD20 is B lymphocyte cell-surface molecule. It is a 33-37 kDa non-glycosylated protein. CD20 is expressed on the surface of B-cells from the pre-B phase, the expression is lost in terminally differentiated plasma cells. CD20 is used as a therapeutical target of B-cell malignancies and autoimmune diseases.

Immunogen: Recombinant protein within human CD20 aa 121-209.

Positive control: Raji cell lysate, Ramos cell lysate, Daudi cell lysate, human spleen tissue, Raji.

Subcellular location: Cell membrane.

Database links: SwissProt: P11836 Human

Recommended Dilutions:

WB	1:10,000
IF-Cell	1:100
IHC-P	1:200
FC	1:1,000
IP	1-2μg/sample

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

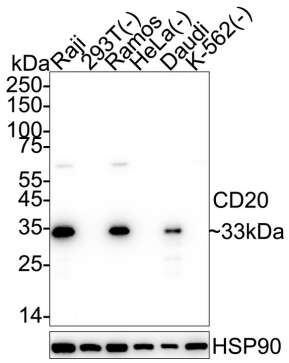

Fig1: Western blot analysis of CD20 on different lysates with Rabbit anti-CD20 antibody (HA751207) at 1/10,000 dilution.

Lane 1: Raji cell lysate
Lane 2: 293T cell lysate (negative)
Lane 3: Ramos cell lysate
Lane 4: HeLa cell lysate (negative)
Lane 5: Daudi cell lysate
Lane 6: K-562 cell lysate (negative)

Lysates/proteins at 20 µg/Lane.

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

Exposure time: 30 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 (HA751207) at 1/10,000 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.

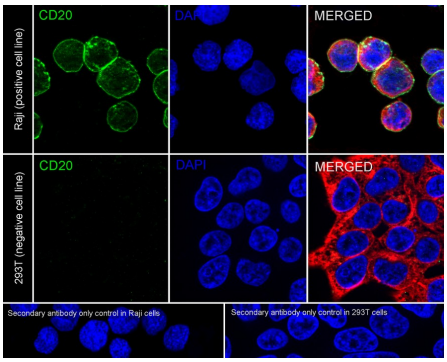

Fig2: Immunocytochemistry analysis of Raji (positive) and 293T (negative) labeling CD20 with Rabbit anti-CD20 antibody (HA751207) at 1/100 dilution.

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-CD20 antibody (HA751207) 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 (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.

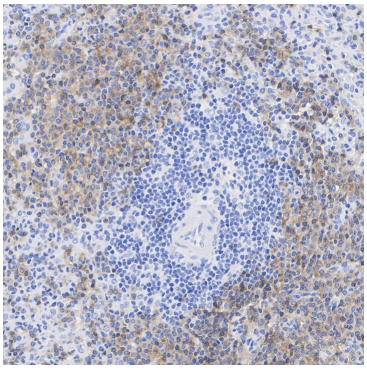

Fig3: Immunohistochemical analysis of paraffin-embedded human spleen tissue with Rabbit anti-CD20 antibody (HA751207) at 1/200 dilution.

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 (HA751207) 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.

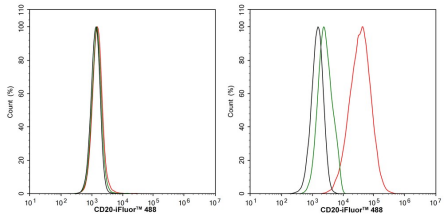

Fig4: Flow cytometric analysis of 293T (left, negative) and Raji (right, positive) cells labeling CD20.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA751207, 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).

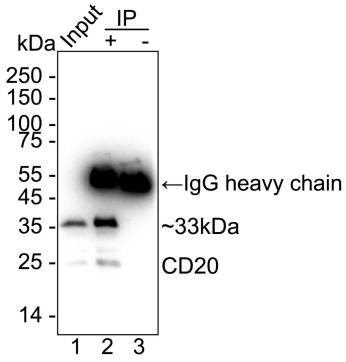

Fig5: CD20 was immunoprecipitated from 0.2 mg Raji cell lysate with HA751207 at 2 µg/25 µl agarose. Western blot was performed from the immunoprecipitate using HA751207 at 1/1,000 dilution. Mouse anti Rabbit IgG heavy chain (Fc) secondary antibody (M1003-7) at 1/100,000 dilution was used for 1 hour at room temperature.

Lane 1: Raji cell lysate (input)
Lane 2: HA751207 IP in Raji cell lysate
Lane 3: Rabbit IgG instead of HA751207 in Raji cell lysate

Blocking/Dilution buffer: 5% NFDM/TBST
Exposure time: 30 seconds; ECL: K1801

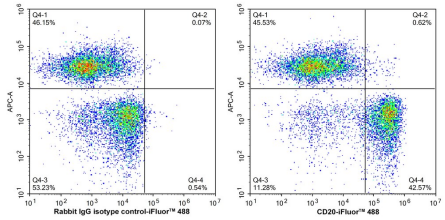

Fig6: Flow cytometric analysis of human PBMC labelling CD20 (HA751207).

Background References

1. de Sèze J et al. Anti-CD20 therapies in multiple sclerosis: From pathology to the clinic. Front Immunol. 2023 Mar
2. Margoni M et al. Anti-CD20 therapies for multiple sclerosis: current status and future perspectives. J Neurol. 2022 Mar