Anti-CD45 Antibody

HA500575

Product Type:	Rabbit polyclonal IgG, primary antibodies
Species reactivity:	Mouse
Applications:	WB, IF-Cell, FC
Molecular Wt:	Predicted band size: 145 kDa

Description: CD45, also known as Protein Tyrosine Phosphatase Receptor Type C (PTPRC), is a member of the protein tyrosine phosphatase (PTP) family. It is a type I transmembrane protein expressed on the surface of all differentiated hematopoietic cells, except erythrocytes and plasma cells. CD45 plays a crucial role in regulating T-cell and B-cell antigen receptor signaling, which is essential for immune cell activation and function. CD45 consists of an extracellular domain, a single transmembrane segment, and two tandem intracytoplasmic catalytic domains. While only one of the cytoplasmic domains has intrinsic phosphatase activity, both domains are necessary for substrate recruitment. CD45 activates Src family kinases, such as Lck, which are required for T-cell receptor (TCR) signaling. It also modulates the activity of other kinases, including LYN and FYN, and suppresses JAK kinases, thereby acting as a negative regulator of cytokine receptor signaling. CD45 is involved in various cellular processes, including cell growth, differentiation, and immune response regulation. Dysregulation of CD45 function can lead to severe combined immunodeficiency and is implicated in autoimmune diseases, cancer, and infectious diseases. Additionally, CD45 exists in multiple isoforms, which are used in immunohistochemistry to differentiate between immune cell types and diagnose lymphomas and carcinomas.

Immunogen:	Recombinant protein within mouse CD45 aa 1-566.
Positive control:	A20 cell lysate, Mouse spleen tissue lysate, A20.
Subcellular location:	Cell membrane, Membrane raft, Synapse.
Database links:	SwissProt: P06800 Mouse

Recommended Dilutions:

WB	1:5,000
IF-Cell	1:250
FC	1:1,000

Storage Buffer:	1*PBS (pH7.4), 0.1% BSA, 40% Glycerol, 0.2% Proclean 950.
Storage Instruction:	Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.
Purity:	Immunogen affinity purified.

Images

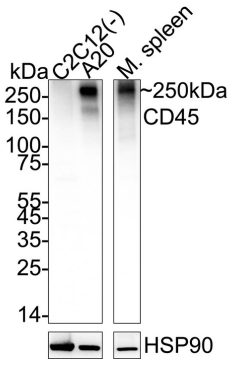

Fig1: Western blot analysis of CD45 on different lysates with Rabbit anti-CD45 antibody (HA500575) at 1/5,000 dilution.

Lane 1: C2C12 cell lysate (negative) (30 µg/Lane)
Lane 2: A20 cell lysate (10 µg/Lane)
Lane 3: Mouse spleen tissue lysate (10 µg/Lane)

Predicted band size: 145 kDa
Observed band size: 250 kDa

Exposure time: 2 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 (HA500575) at 1/5,000 dilution was used in primary antibody dilution (K1803) at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

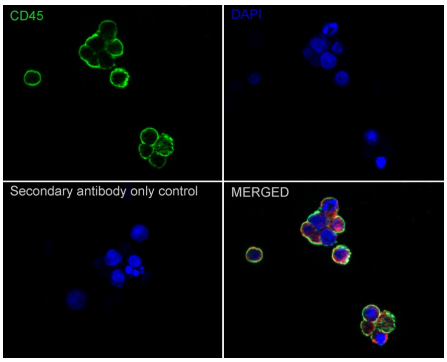

Fig2: Immunocytochemistry analysis of A20 cells labeling CD45 with Rabbit anti-CD45 antibody (HA500575) at 1/250 dilution.

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-CD45 antibody (HA500575) 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.

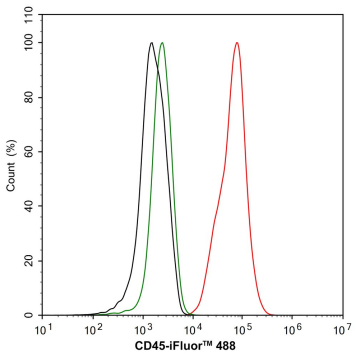

Fig3: Flow cytometric analysis of A20 cells labeling CD45.

Cells were washed twice with cold PBS and resuspend. Then stained with the primary antibody (HA500575, 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).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Katagiri T, et al. CD45 negatively regulates lyn activity by dephosphorylating both positive and negative regulatory tyrosine residues in immature B cells. J Immunol. 1999, Aug. Holmes N. CD45: all is not yet crystal clear. Immunology. 2006, Feb.