

# Anti-CD45 Antibody Picoband™ (monoclonal, 3H6)

Catalog # ABO15074

## Product Information

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| <b>Application</b>       | WB, IHC, IF, FC                                                                                                                       |
| <b>Primary Accession</b> | <a href="#">P08575</a>                                                                                                                |
| <b>Host</b>              | Mouse                                                                                                                                 |
| <b>Isotype</b>           | Mouse IgG1                                                                                                                            |
| <b>Reactivity</b>        | Human                                                                                                                                 |
| <b>Clonality</b>         | Monoclonal                                                                                                                            |
| <b>Format</b>            | Lyophilized                                                                                                                           |
| <b>Description</b>       | Anti-CD45 Antibody Picoband™ (monoclonal, 3H6) . Tested in Flow Cytometry, IF, IHC, WB applications. This antibody reacts with Human. |
| <b>Reconstitution</b>    | Adding 0.2 ml of distilled water will yield a concentration of 500 µg/ml.                                                             |

## Additional Information

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| <b>Gene ID</b>             | 5788                                                                                                                                                                                                    |
| <b>Other Names</b>         | Receptor-type tyrosine-protein phosphatase C, 3.1.3.48, Leukocyte common antigen, L-CA, T200, CD45, PTPRC ( <a href="#">HGNC:9666</a> ), CD45                                                           |
| <b>Calculated MW</b>       | 147486                                                                                                                                                                                                  |
| <b>Application Details</b> | Western blot, 0.25-0.5 µg/ml, Human<br>Immunohistochemistry(Paraffin-embedded Section), 2-5 µg/ml, Human<br>Immunofluorescence, 5 µg/ml, Human<br>Flow Cytometry, 1-3 µg/1x10 <sup>6</sup> cells, Human |
| <b>Source</b>              | Eukaryota                                                                                                                                                                                               |
| <b>Contents</b>            | Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> .                                                                                                            |
| <b>Clone Names</b>         | Clone: 3H6                                                                                                                                                                                              |
| <b>Immunogen</b>           | A synthetic peptide corresponding to a sequence at the C-terminus of human CD45, different from the related mouse sequence by eight amino acids, and from the related rat sequence by ten amino acids.  |
| <b>Purification</b>        | Immunogen affinity purified.                                                                                                                                                                            |
| <b>Storage</b>             | At -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freezing and thawing.       |

## Protein Information

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| <b>Name</b>              | PTPRC ( <a href="#">HGNC:9666</a> )                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                              |
| <b>Synonyms</b>          | CD45                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                             |
| <b>Function</b>          | Protein tyrosine-protein phosphatase required for T-cell activation through the antigen receptor (PubMed: <a href="#">35767951</a> ). Acts as a positive regulator of T-cell coactivation upon binding to DPP4. The first PTPase domain has enzymatic activity, while the second one seems to affect the substrate specificity of the first one. Upon T-cell activation, recruits and dephosphorylates SKAP1 and FYN. Dephosphorylates LYN, and thereby modulates LYN activity (By similarity). Interacts with CLEC10A at antigen presenting cell-T cell contact; CLEC10A on immature dendritic cells recognizes Tn antigen- carrying PTPRC/CD45 receptor on effector T cells and modulates T cell activation threshold to limit autoreactivity. |
| <b>Cellular Location</b> | Cell membrane; Single-pass type I membrane protein. Membrane raft. Synapse. Note=Colocalized with DPP4 in membrane rafts.                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                                        |
| <b>Tissue Location</b>   | Isoform 1: Detected in thymocytes. Isoform 2: Detected in thymocytes. Isoform 3: Detected in thymocytes. Isoform 4: Not detected in thymocytes. Isoform 5: Detected in thymocytes. Isoform 6: Not detected in thymocytes. Isoform 7: Detected in thymocytes Isoform 8: Not detected in thymocytes.                                                                                                                                                                                                                                                                                                                                                                                                                                               |

## Background

CD45 (Cluster of Differentiation 45), also known as PTPRC, LCA or CD45R, is an enzyme that, in humans, is encoded by the PTPRC gene. It is a member of the protein tyrosine phosphatase (PTP) family. CD45 is a major high molecular mass leukocyte cell surface molecule which is also an integral membrane protein tyrosine phosphatase. The cytogenetic location of CD45 is 1q31.3-q32.1. This gene is especially a prototype for transmembrane protein-tyrosine phosphatase (PTP). Targeted disruption of the CD45 gene leads to enhanced cytokine and interferon receptor-mediated activation of JAKs and STAT proteins. In vitro, CD45 directly dephosphorylates and binds to JAKs. Functionally, CD45 negatively regulates interleukin-3-mediated cellular proliferation, erythropoietin-dependent hematopoiesis, and antiviral responses in vitro and in vivo. In addition, CD45 has been best studied in T cells, where it determines T cell receptor signaling thresholds. CD45 is moved into or out of the immunological synapse (IS) membrane microdomain depending on the relative influence of interaction with the extracellular galectin lattice or the intracellular actin cytoskeleton. Galectin interaction can be finetuned by varying usage of the heavily Oglycosylated spliced regions and sialylation of Nlinked carbohydrates.

## Images

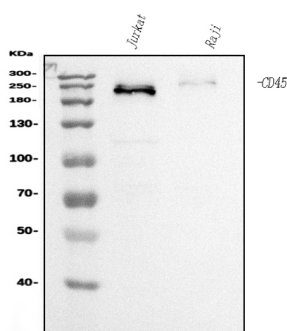


Figure 1. Western blot analysis of CD45 using anti-CD45 antibody (M00555-6).

Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions.

Lane 1: human Jurkat whole cell lysates,

Lane 2: human Raji whole cell lysates.

After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-CD45 antigen affinity purified monoclonal antibody (Catalog # M00555-6) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary

antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for CD45 at approximately 220 kDa. The expected band size for CD45 is at 220 kDa.

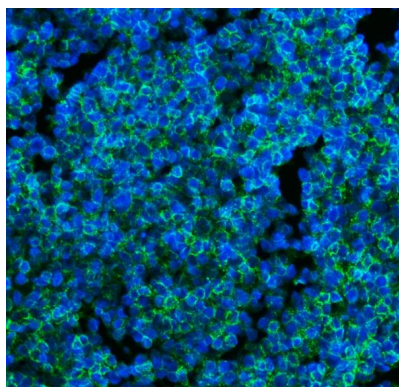


Figure 2. IF analysis of CD45 using anti-CD45 antibody (M00555-6).

CD45 was detected in a paraffin-embedded section of human tonsil tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 5 µg/mL mouse anti-CD45 Antibody (M00555-6) overnight at 4°C. Biotin conjugated goat anti-mouse IgG (BA1001) was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using DyLight®488 Conjugated Avidin (BA1128). The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

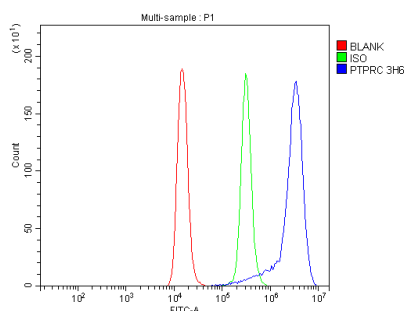


Figure 3. Flow Cytometry analysis of Raji cells using anti-CD45 antibody (M00555-6).

Overlay histogram showing Raji cells stained with M00555-6 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-CD45 Antibody (M00555-6, 1 µg/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 µg/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 µg/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample (Red line) was also used as a control.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.