abcam

Product datasheet

Anti-CD45 antibody [F10-89-4] - Hematopoietic Stem Cell Marker ab30470

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Overview

Product name Anti-CD45 antibody [F10-89-4] - Hematopoietic Stem Cell Marker

Description Mouse monoclonal [F10-89-4] to CD45 - Hematopoietic Stem Cell Marker

Host species Mouse

Specificity Clone F10-89-4 reacts with all forms of CD45 expressed by all haematopoietic cells, except

erythrocytes, having a higher level of expression on lymphocytes than on granulocytes.

Tested applications Suitable for: ICC/IF, Flow Cyt, IHC-P

Species reactivity Reacts with: Human

Immunogen Tissue, cells or virus corresponding to Human CD45. Human T lymphocytes.

Positive control IHC-P: Human tonsil (normal) tissue. ICC/IF: Jurkat cells. Flow Cyt: Jurkat cells, whole blood.

General notes

This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Purity Protein G purified

Primary antibody notes Clone F10-89-4 reacts with all forms of CD45 expressed by all haematopoietic cells, except

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erythrocytes, having a higher level of expression on lymphocytes than on granulocytes.

ClonalityMonoclonalClone numberF10-89-4MyelomaNS1IsotypeIgG2a

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab30470 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF	**** <u>(1)</u>	Use a concentration of 5 - 10 µg/ml.
Flow Cyt		1/10 - 1/50. ab170191 - Mouse monoclonal lgG2a, is suitable for use as an isotype control with this antibody.
IHC-P		Use a concentration of 1 - 5 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Target

Function

Protein tyrosine-protein phosphatase required for T-cell activation through the antigen receptor. 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.

Involvement in disease

Defects in PTPRC are a cause of severe combined immunodeficiency autosomal recessive T-cell-negative/B-cell-positive/NK-cell-positive (T(-)B(+)NK(+) SCID) [MIM:608971]. A form of severe combined immunodeficiency (SCID), a genetically and clinically heterogeneous group of rare congenital disorders characterized by impairment of both humoral and cell-mediated immunity, leukopenia, and low or absent antibody levels. Patients present in infancy recurrent, persistent infections by opportunistic organisms. The common characteristic of all types of SCID is absence of T-cell-mediated cellular immunity due to a defect in T-cell development.

Genetic variations in PTPRC are involved in multiple sclerosis susceptibility (MS) [MIM:126200]. MS is a neurodegenerative disorder characterized by the gradual accumulation of focal plaques of demyelination particularly in the periventricular areas of the brain. Peripheral nerves are not affected. Onset usually in third or fourth decade with intermittent progression over an extended period. The cause is still uncertain.

Sequence similarities

Belongs to the protein-tyrosine phosphatase family. Receptor class 1/6 subfamily.

Contains 2 fibronectin type-III domains.

Contains 2 tyrosine-protein phosphatase domains.

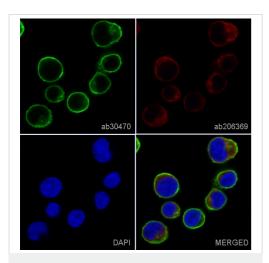
Domain

The first PTPase domain interacts with SKAP1.

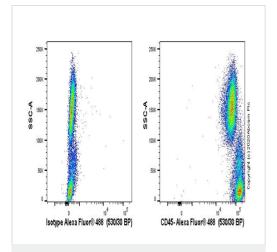
Post-translational modifications

Heavily N- and O-glycosylated.

Images



Immunocytochemistry/ Immunofluorescence - Anti-CD45 antibody [F10-89-4] - Hematopoietic Stem Cell Marker (ab30470)



Flow Cytometry - Anti-CD45 antibody [F10-89-4] - Hematopoietic Stem Cell Marker (ab30470)

ab30470 staining CD45 in Jurkat (Human T cell leukemia cell line from peripheral blood) cells. The cells were fixed with 4% formaldehyde (10 minutes), then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1 hour. The cells were then incubated overnight at +4°C with ab30470 at 5 μ g/ml (shown in green) and <u>ab206369</u>, Rabbit monoclonal to beta Tubulin (Alexa Fluor® 594), at 1/250 dilution (shown in red). This was followed by an incubation at room temperature for 1 hour with <u>ab150117</u>, Goat Anti-Mouse IgG H&L (Alexa Fluor® 488) preadsorbed, at 1 μ g/ml (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

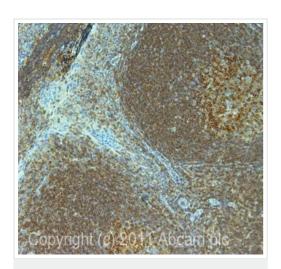
Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).

This product also gave a positive signal under the same testing conditions in Jurkat cells fixed with 80% methanol (5 minutes).

Flow cytometry staining of human whole blood with ab30470 (right) or mouse $\lg G2a\kappa$; (ab18413) isotype (left). Red blood cells of 200 μL blood were lysed, then cells were incubated for 30 min on ice in 1x PBS containing 10 μ g/ml human $\lg G$ and 10 % normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody (ab30470) or mouse $\lg G2a\kappa$; (ab18413) isotype (1x10⁶ in 100 μL at 0.2 μ g/ml) for 30 min on ice.

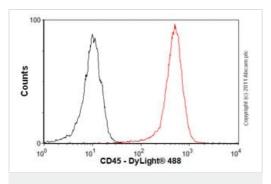
The secondary antibody Goat anti-mouse IgG H&L (Alexa Fluor [®] 488, pre-adsorbed) (<u>ab150117</u>) was used at 1/2000 dilution for 30 min on ice.

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 530/30 bandpass filter. Events were gated on alive cells.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD45 antibody [F10-89-4] - Hematopoietic Stem Cell Marker (ab30470)

IHC image of CD45 staining in human tonsil formalin fixed paraffin embedded tissue section, performed on a Leica Bond TM system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6, epitope retrieval solution 1) for 20 minutes. The section was then incubated with ab30470, 1 μ g/ml, for 15 minutes at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.



Flow Cytometry - Anti-CD45 antibody [F10-89-4] - Hematopoietic Stem Cell Marker (ab30470)

Overlay histogram showing Jurkat (Human T cell leukemia cell line from peripheral blood) cells stained with ab30470 (red line). The cells were fixed with 4% paraformaldehyde (10 minutes) and incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab30470, 1 μ g/1 x 10⁶ cells) for 30 minutes at 22°C. The secondary antibody used was DyLight[®] 488 goat anti-mouse lgG (H+L) (ab96879) at 1/500 dilution for 30 minutes at 22°C. Isotype control antibody (black line) was mouse lgG2a [ICIGG2A] (ab91361, 1 μ g/1 x 10⁶ cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in Jurkat cells fixed with 80% methanol (5 minutes) used under the same conditions.

Please note that Abcam do not have any data for use of this antibody on non-fixed cells. We welcome any customer feedback.

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