abcam

Product datasheet

# Anti-CD45 antibody ab10558

58 Abreviews 375 References 8 图像

## 概述

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|---|---|
| **产品名称** | Anti-CD45抗体 |
| **描述** | 兔多克隆抗体to CD45 |
| **宿主** | Rabbit |
| **经测试应用** | **适用于:** WB, IHC-P, Flow Cyt (Intra) |
| **种属反应性** | **与反应:** Mouse, Rat, Human<br>**预测可用于:** Pig, Rhesus monkey |
| **免疫原** | Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. |
| **常规说明** | 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.<br>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 |

## 性能

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| **形式** | Liquid |
| **存放说明** | Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw cycles. |
| **存储溶液** | pH: 7.40<br>Preservative: 0.02% Sodium azide<br>Constituents: PBS, 1% BSA |
| **纯度** | Immunogen affinity purified |
| **克隆** | 多克隆 |
| **同种型** | IgG |

## 应用

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| **The Abpromise guarantee** | Abpromise™承诺保证使用ab10558于以下的经测试应用 |

“应用说明”部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

| 应用 | Ab评论 | 说明 |
|---|---|---|
| **WB** | ★★★★☆ (9) | 1/500. Detects a band of approximately 190 kDa (predicted molecular weight: 147 kDa). |
| **IHC-P** | ★★★★☆ (37) | Use a concentration of 0.5 - 5 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. |
| **Flow Cyt (Intra)** | | Use 1µg for $10^6$ cells.<br>**ab171870** - Rabbit polyclonal IgG, is suitable for use as an isotype control with this antibody. |

## 靶标

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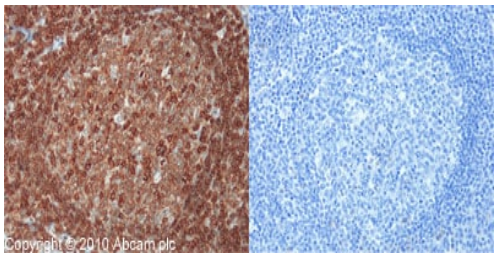
|---|---|
| **功能** | Protein tyrosine-protein phosphatase required for T-cell activation through the antigen receptor. Acts as a positive regulator of T-cell coactivation upon binding to DPP4. The first PTPase domain has enzymatic activity, while the second one seems to affect the substrate specificity of the first one. Upon T-cell activation, recruits and dephosphorylates SKAP1 and FYN. |
| **疾病相关** | 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.<br>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. |
| **序列相似性** | Belongs to the protein-tyrosine phosphatase family. Receptor class 1/6 subfamily.<br>Contains 2 fibronectin type-III domains.<br>Contains 2 tyrosine-protein phosphatase domains. |
| **结构域** | The first PTPase domain interacts with SKAP1. |
| **翻译后修饰** | Heavily N- and O-glycosylated. |
| **细胞定位** | Membrane. Membrane raft. Colocalized with DPP4 in membrane rafts. |

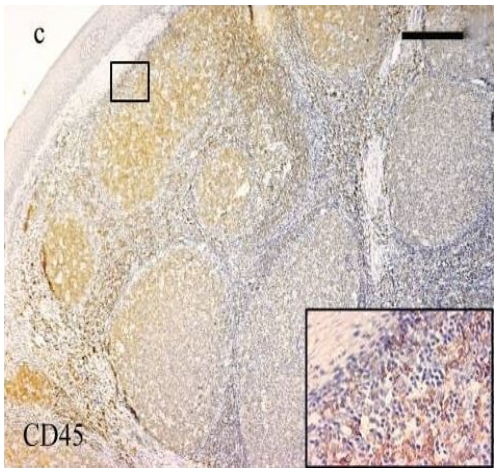
## 图片




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD45 antibody (ab10558)

ab10558 (1:40) staining CD45 in paraffin-embedded human tonsil (left panel) using an automated system (Ventana Discovery). Right-hand panel shows negative control (no primary antibody).
Using this protocol there is strong membrane staining of B cells in the germinal centres and mantle zone of the follicles and scattered cells of the interfollicular areas (paracortical T and B cells). There is a mild to moderate degree of cytoplasmic staining associated with the membrane staining in these specific cells.

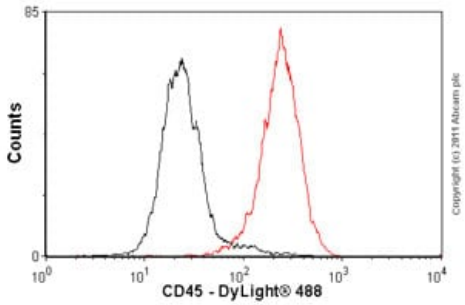
Sections were rehydrated and antigen retrieved in CC1 Cell Conditioning Buffer using Ventana Extended Retrieval programme. Slides were blocked in 3% $H_2O_2$ /4 min/ 37°C and incubated with ab10558 (1:40 dilution / 1 hour/ 37°C). Sections then blocked (4mins/ 37°C) and incubated with Dako swine anti-rabbit antibody (1:50, 28 min/ 37°C). Staining was amplified and detected by incubation with Ventana Streptavidin ABC (HRP-DAB) system (16 min/ 37°C) before being counterstained with hematoxylin.




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD45 antibody (ab10558)



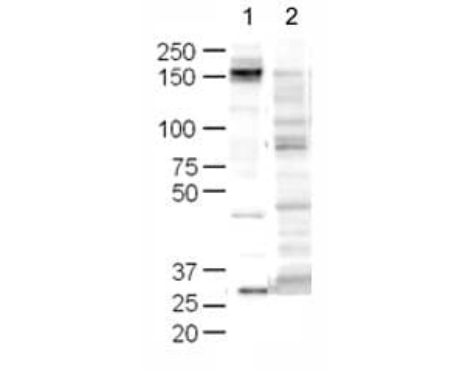
ab10558 staining CD45 in Human tonsil tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Negative control is shown in panel. Blocking was with horse serum (1/75) for 1 hour at room temperature. Samples were incubated with primary antibody (1/10) overnight at 4°C. A Biotin-conjugated Horse anti-mouse polyclonal (1/200) was used as the secondary antibody.




Flow Cytometry (Intracellular) - Anti-CD45 antibody (ab10558)

Overlay histogram showing Jurkat cells stained with ab10558 (red line). The cells were fixed with 80% methanol (5 min) 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 (ab10558, 1µg/1x$10^6$ cells) for 30 min at 22ºC. The secondary antibody used was **DyLight® 488 goat anti-rabbit IgG (H+L)** (**ab96899**) at 1/1000 dilution for 30 min at 22ºC. Isotype control antibody (black line) was rabbit IgG (1µg/1x$10^6$ cells) used under the same conditions. Acquisition of >5,000 events was performed.

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




Western blot - Anti-CD45 antibody (ab10558)

**All lanes :** Anti-CD45 antibody (ab10558) at 1/500 dilution

**Lane 1 :** Jurkat Whole Cell Lysate
**Lane 2 :** Jurkat Whole Cell Lysate with Human CD45 peptide (**ab17553**)

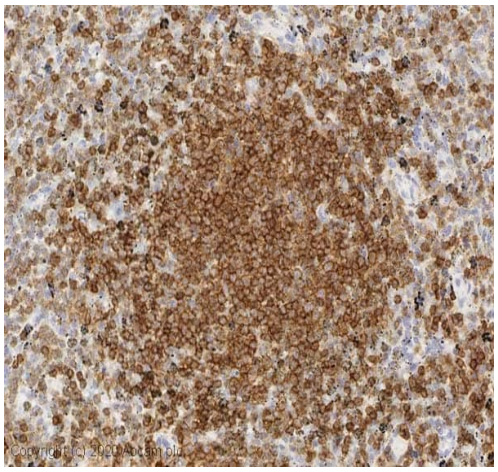
Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (**ab6721**) at 1/5000 dilution

**Predicted band size:** 147 kDa

**Exposure time:** 3 minutes

**Secondary antibody - goat anti-rabbit H&L (HRP) (ab6721)**

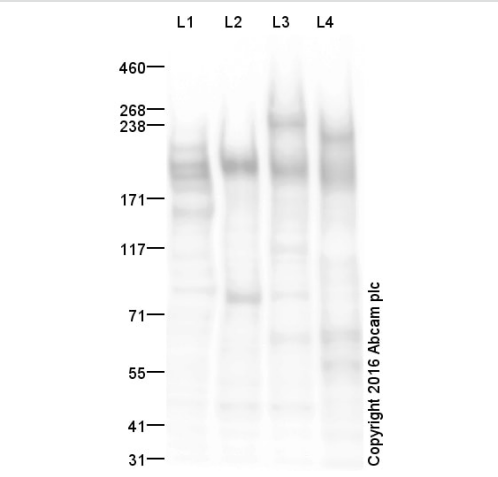


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD45 antibody (ab10558)

IHC image of CD45 antibody staining in a section of formalin-fixed paraffin-embedded normal human spleen* performed on a Leica BOND™ system using the standard protocol. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab10558, 1ug/ml, for 15 mins 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.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

**Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre*




Western blot - Anti-CD45 antibody (ab10558)

**All lanes :** Anti-CD45 antibody (ab10558) at 1 µg/ml

**Lane 1 :** Jurkat (Human) Whole Cell Lysate
**Lane 2 :** RAW 264.7 (Mouse leukaemic monocyte macrophage cell line) Whole Cell Lysate
**Lane 3 :** Spleen (Mouse) Tissue Lysate
**Lane 4 :** Spleen (Rat) Tissue Lysate

Lysates/proteins at 20 µg per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) at 1/50000 dilution

Developed using the ECL technique.

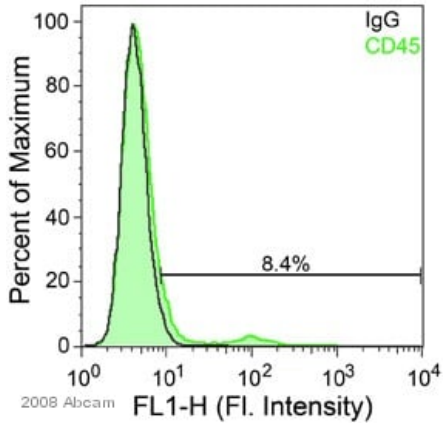
Performed under reducing conditions.

**Predicted band size:** 147 kDa
**Observed band size:** 190 kDa
**Additional bands at:** 230 kDa. We are unsure as to the identity of these extra bands.

**Exposure time:** 1 minute

This blot was produced using a 3-8% Tris Acetate gel under the TA buffer system. The gel was run at 150V for 60 minutes before being transferred onto a Nitrocellulose membrane at 30V for 70 minutes. The membrane was then blocked for an hour using 2% Bovine Serum Albumin before being incubated with ab10558 overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP, and visualised using ECL development solution **ab133406**.

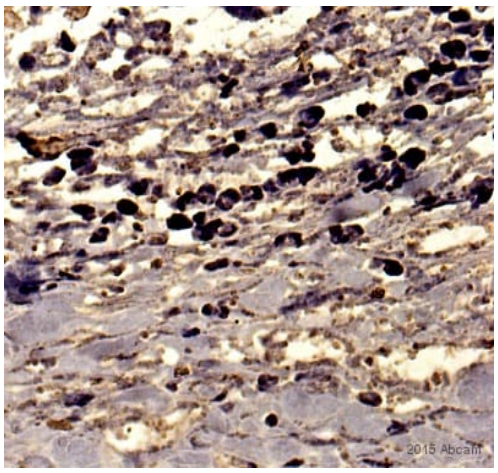
CD45 contains a number of potential glycosylation sites (SwissProt) which may explain its migration at a higher molecular weight than predicted.




Flow Cytometry (Intracellular) - Anti-CD45 antibody (ab10558)

This image is courtesy of an abreview submitted by Kirk Mcmanu.

Asynchronous KM-H2 cells were pelleted and labeled by indirect immunofluorescence. Cells were stained with ab10558 (1/200) for 30min at 4'C, washed and then stained with goat anti-rabbit alexafluor 488 (1/200). Forward/Side scatter were used to eliminate cellular debris. The accompanying marker was applied such that only 2% of the IgG control was positive Based on the accompanying image, approximately 8.4% of cells exhibited positive staining for anti-CD45. Since KM-H2 are known to have low levels of CD45 transcripts they are expected to have low levels of CD45, which is reflected in the ~8%. This image is from an Abreview.




Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CD45 antibody (ab10558)
This image is courtesy of an anonymous abreview.

Immunohistochemical analysis of formaldehyde fixed human cephalic sections. Primary antibody ab10558 to CD45 incubated at a concentration of 1/100 for 4°C for 18 hours. Secondary antibody used was a goat anti-rabbit congugated to biotin at a 1/200 dilution. Blocking was done with serum at a 10% concentration for 1 hour at 25°C.

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