abcam

Product datasheet

Anti-DAP12 antibody [EPR5173] ab124834

Recombinant RabMAb

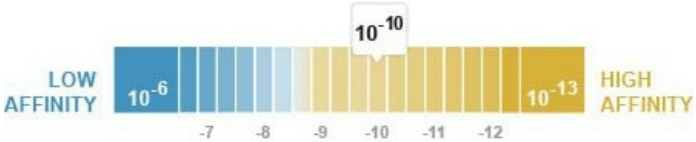
★★★★★ 1 Abreviews 6 References 6 Images

Overview

Product name	Anti-DAP12 antibody [EPR5173]
Description	Rabbit monoclonal [EPR5173] to DAP12
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), ICC, WB **Unsuitable for:** IHC-P or IP
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide within Human DAP12 aa 50-150. The exact sequence is proprietary. Database link: **O43914**
Positive control	Peripheral blood leukocytes, Human placenta and peripheral blood mononuclear cell lysates.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
Dissociation constant (K_D)	$K_D = 1.86 \times 10^{-10}$ M



Learn more about K_D

Storage buffer	pH: 7.20 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
Purity	Tissue culture supernatant
Purification notes	Not purified; provided in cell supernatant and storage buffer.
Clonality	Monoclonal
Clone number	EPR5173
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab124834 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
Flow Cyt (Intra)		1/100. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
ICC		1/250 - 1/500.
WB		1/1000 - 1/10000. Predicted molecular weight: 12 kDa.

Application notes Is unsuitable for IHC-P or IP.

Target

Function	Non-covalently associates with activating receptors of the CD300 family. Cross-linking of CD300-TYROBP complexes results in cellular activation.
Tissue specificity	Expressed at low levels in the early development of the hematopoietic system and in the promonocytic stage and at high levels in mature monocytes. Expressed in hematological cells and tissues such as peripheral blood leukocytes and spleen. Also found in bone marrow, lymph nodes, placenta, lung and liver. Expressed at lower levels in different parts of the brain especially in the basal ganglia and corpus callosum.
Involvement in disease	Defects in TYROBP are a cause of polycystic lipomembranous osteodysplasia with sclerosing leukoencephalopathy (PLOSL) [MIM:221770]; also called presenile dementia with bone cysts or Nasu-Hakola disease (NHD). PLOSL is a recessively inherited disease characterized by a combination of psychotic symptoms rapidly progressing to presenile dementia and bone cysts restricted to wrists and ankles. PLOSL has a global distribution, although most of the patients have been diagnosed in Finland and Japan, with an estimated population prevalence of 2x10(-6) in the Finns.
Sequence similarities	Belongs to the TYROBP family.
Post-translational	Tyrosine phosphorylated.

modifications

Cellular localization Membrane.

Images

Western blot - Anti-DAP12 antibody [EPR5173] (ab124834)

All lanes : Anti-DAP12 antibody [EPR5173] (ab124834) at 1/1000 dilution

Lane 1 : Wild-type THP-1 cell lysate
Lane 2 : TYROBP CRISPR-Cas9 edited THP-1 cell lysate

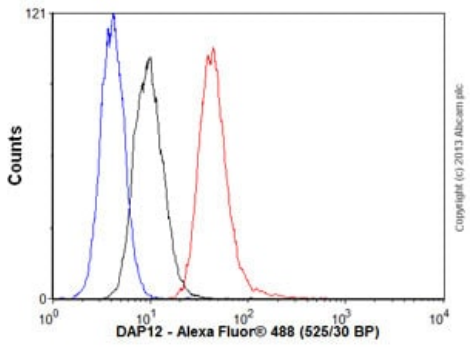
Lysates/proteins at 40 µg per lane.

Performed under reducing conditions.

Predicted band size: 12 kDa
Observed band size: 14 kDa

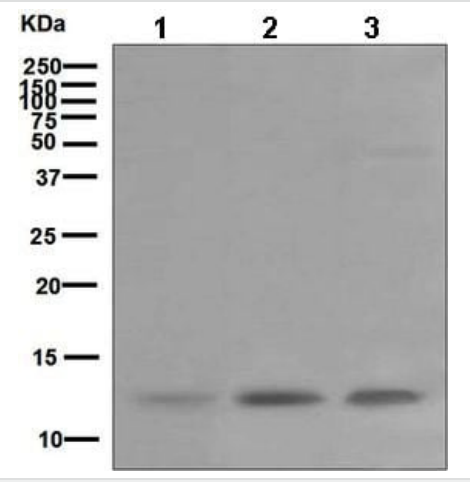
False colour image of Western blot: Anti-DAP12 antibody [EPR5173] staining at 1/1000 dilution, shown in black; Mouse anti-GAPDH antibody [6C5] (**ab8245**) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab124834 was shown to bind specifically to DAP12. A band was observed at 14 kDa in wild-type THP-1 cell lysates with no signal observed at this size in TYROBP CRISPR-Cas9 edited cell line **ab273726** (CRISPR-Cas9 edited cell lysate **ab273774**). The band observed in the CRISPR-Cas9 edited lysate lane below 14 kDa is likely to represent a truncated form of DAP12. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and TYROBP CRISPR-Cas9 edited THP-1 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3% milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4°C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times before development with Optiblot (ECL reagent **ab133456**) and imaged with 20 minutes exposure time. Secondary antibodies used were HRP conjugated Goat anti-Rabbit (H+L) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) at 1/20000

dilution.



Flow Cytometry (Intracellular) - Anti-DAP12 antibody [EPR5173] (ab124834)

Overlay histogram showing THP1 cells stained with ab124834 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% human serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab124834, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



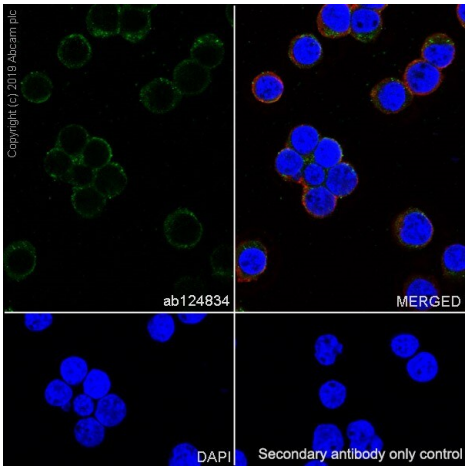
Western blot - Anti-DAP12 antibody [EPR5173] (ab124834)

All lanes : Anti-DAP12 antibody [EPR5173] (ab124834) at 1/1000 dilution

Lane 1 : Peripheral blood leukocytes lysates
Lane 2 : Human placenta lysates
Lane 3 : Peripheral blood mononuclear lysates

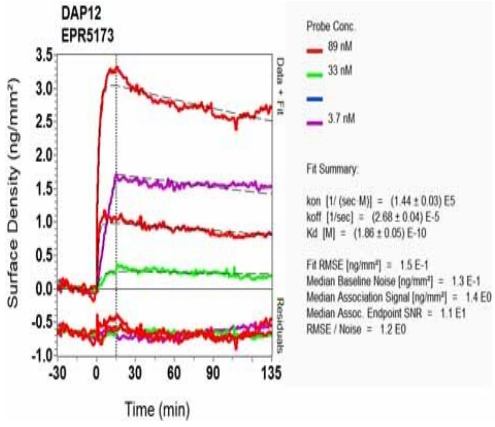
Lysates/proteins at 10 µg per lane.

Predicted band size: 12 kDa



Immunocytochemistry - Anti-DAP12 antibody [EPR5173] (ab124834)

Immunocytochemistry analysis of Jurkat (human T cell leukemia T lymphocyte) labeling DAP12 with purified ab124834 at 1/100 dilution (10 µg/ml). Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/1000 (2 µg/ml) was used as the secondary antibody. **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.7 µg/ml) was used as counterstain. Nuclei were stained blue with DAPI. Negative control: PBS instead of the primary antibody.

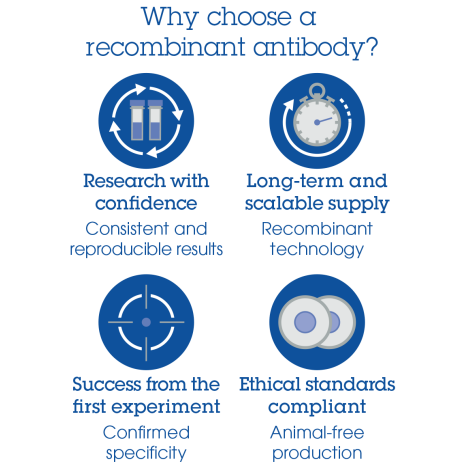


OI-RD Scanning - Anti-DAP12 antibody [EPR5173] (ab124834)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D



Anti-DAP12 antibody [EPR5173] (ab124834)

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