abcam

Product datasheet

Anti-CD33 antibody [EPR4423] - Low endotoxin, Azide free ab215383

KO VALIDATED Recombinant RabMAb

4 Images

Overview

Product name	Anti-CD33 antibody [EPR4423] - Low endotoxin, Azide free
Description	Rabbit monoclonal [EPR4423] to CD33 - Low endotoxin, Azide free
Host species	Rabbit
Tested applications	**Suitable for:** WB, IP
Species reactivity	**Reacts with:** Human
Immunogen	Full length protein. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: THP1 cell lysate, U937 cell lysate, HL-60 cell lysate, HepG2 whole cell lysate (**ab7900**), HeLa whole cell lysate (**ab150035**) Flow Cyt: THP1 cells
General notes	ab215383 is the carrier-free version of **ab134115**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. 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**. Our **Low endotoxin, azide-free formats** have low endotoxin level (≤ 1 EU/ml, determined by the LAL assay) and are free from azide, to achieve consistent experimental results in functional assays.

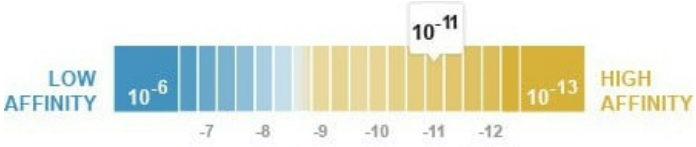
Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C. Do Not Freeze.

Dissociation constant (K_D) $K_D = 1.76 \times 10^{-11}$ M



Learn more about K_D

Storage buffer pH: 7.2
Constituent: PBS

Carrier free Yes

Purity Protein A purified

Clonality Monoclonal

Clone number EPR4423

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab215383 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
WB		Use at an assay dependent concentration. Detects a band of approximately 67-75 kDa (predicted molecular weight: 40 kDa).
IP		Use at an assay dependent concentration.

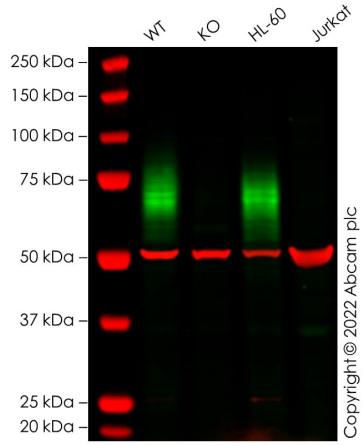
Target

Function Putative adhesion molecule of myelomonocytic-derived cells that mediates sialic-acid dependent binding to cells. Preferentially binds to alpha-2,6-linked sialic acid. The sialic acid recognition site may be masked by cis interactions with sialic acids on the same cell surface. In the immune response, may act as an inhibitory receptor upon ligand induced tyrosine phosphorylation by recruiting cytoplasmic phosphatase(s) via their SH2 domain(s) that block signal transduction through dephosphorylation of signaling molecules. Induces apoptosis in acute myeloid leukemia (in vitro).

Tissue specificity Monocytic/myeloid lineage cells.

Sequence similarities	Belongs to the immunoglobulin superfamily. SIGLEC (sialic acid binding Ig-like lectin) family. Contains 1 Ig-like C2-type (immunoglobulin-like) domain. Contains 1 Ig-like V-type (immunoglobulin-like) domain.
Domain	Contains 2 copies of a cytoplasmic motif that is referred to as the immunoreceptor tyrosine-based inhibitor motif (ITIM). This motif is involved in modulation of cellular responses. The phosphorylated ITIM motif can bind the SH2 domain of several SH2-containing phosphatases.
Post-translational modifications	Phosphorylation of Tyr-340 is involved in binding to PTPN6 and PTPN11. Phosphorylation of Tyr-358 is involved in binding to PTPN6.
Cellular localization	Cell membrane.

Images



Western blot - Anti-CD33 antibody [EPR4423] - Low endotoxin, Azide free (ab215383)

All lanes : Anti-CD33 antibody [EPR4423] (**ab134115**) at 1/1000 dilution

Lane 1 : Wild-type THP-1 cell lysate
Lane 2 : CD33 knockout THP-1 cell lysate
Lane 3 : HL-60 cell lysate
Lane 4 : Jurkat cell lysate

Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

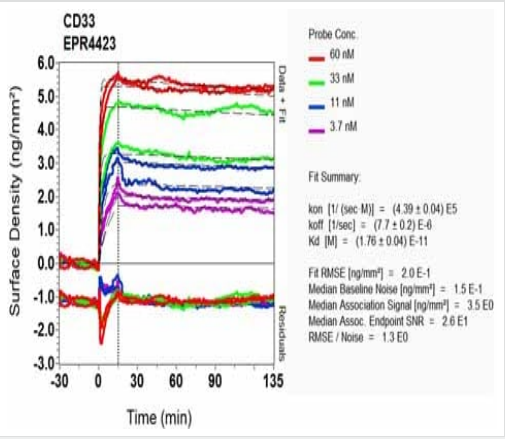
Performed under reducing conditions.

Predicted band size: 40 kDa
Observed band size: 60-80 kDa

False colour image of Western blot: Anti-CD33 antibody [EPR4423] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] (**ab7291**) loading control staining at 1/20000 dilution, shown in red. In Western blot, **ab134115** was shown to bind specifically to CD33. A band was observed at 60-80 kDa in wild-type THP-1 cell lysates with no signal observed at this size in CD33 knockout cell line **ab273831** (knockout cell lysate **ab273785**). To generate this image, wild-type and CD33 knockout 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 then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab134115**).

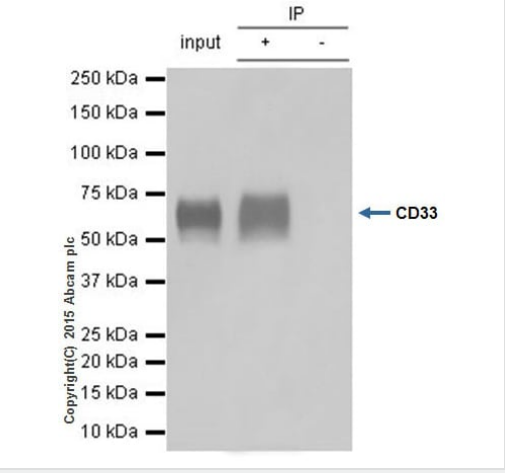


OI-RD Scanning - Anti-CD33 antibody [EPR4423] - Low endotoxin, Azide free (ab215383)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D



Immunoprecipitation - Anti-CD33 antibody [EPR4423] - Low endotoxin, Azide free (ab215383)

This IP data was generated using the same anti-CD33 antibody clone, EPR4423, in a different buffer formulation (cat# **ab134115**).

ab134115 (purified) at 1/60 immunoprecipitating CD33 in 10 µg THP-1 (Lanes 1 and 2, observed at 67-75 kDa). Lane 3 - PBS. For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10,000 dilution. Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST

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