# abcam

# Product datasheet

# Anti-CD33 antibody [EPR4423] - BSA and Azide free ab214439



Recombinant

RabMAb

★★★★★ 1 Abreviews 4 Images

#### Overview

Product name Anti-CD33 antibody [EPR4423] - BSA and Azide free

**Description** Rabbit monoclonal [EPR4423] to CD33 - BSA and Azide free

Host species Rabbit

Tested applications Suitable for: WB, IP, Flow Cyt

Unsuitable for: IHC-P

Species reactivity Reacts with: Human

**Immunogen** Synthetic peptide. 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** ab214439 is the carrier-free version of **ab134115**.

Our <u>carrier-free</u> 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 compatible with the Maxpar $^{(\!0\!)}$  Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar $^{(\!0\!)}$  is a trademark of Fluidigm Canada Inc.

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

1

monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

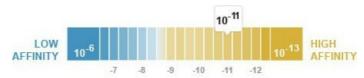
Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

#### **Properties**

Form Liquid

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

**Dissociation constant (K<sub>D</sub>)**  $K_D = 1.76 \times 10^{-11} M$ 



Learn more about KD

Storage buffer pH: 7.2

Constituent: PBS

Carrier free Yes

Purity Protein A purified

ClonalityMonoclonalClone numberEPR4423

**Isotype** IgG

## **Applications**

### The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab214439 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.
Flow Cyt	*****(1)	Use at an assay dependent concentration.

**Application notes** Is unsuitable for IHC-P.

#### **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 lg-like lectin) family.

Contains 1 lg-like C2-type (immunoglobulin-like) domain. Contains 1 lg-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.

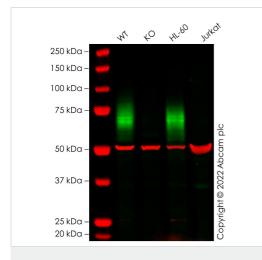
Post-translational modifications

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] - BSA and Azide free (ab214439)

**All lanes :** Anti-CD33 antibody [EPR4423] (<u>ab134115</u>) 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<sup>®</sup> 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).

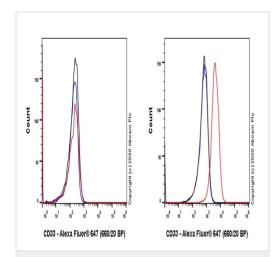
Flow Cytometry analysis of Jurkat (Human T cell leukemia T lymphocyte, Left) / THP1 (Human monocytic leukemia monocyte, Right) labeling CD33 with <u>ab134115</u> at 1/500 dilution (red). Goat anti rabbit lgG (Alexa Fluor<sup>®</sup> 647, <u>ab150079</u>) at 1/2000 dilution was used as the secondary antibody. Rabbit monoclonal lgG (<u>ab172730</u>) was used as isotype control (Black). Unlabelled control: Cell without incubation with primary antibody and secondary antibody (Blue).

Negative control: Jurkat. Gated on viable cells.

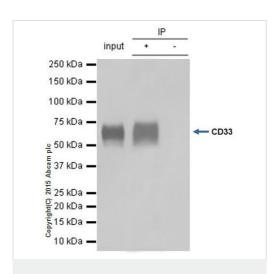
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab134115).

<u>ab134115</u> (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) (<u>ab131366</u>), was used for detection at 1/10,000 dilution. Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST.

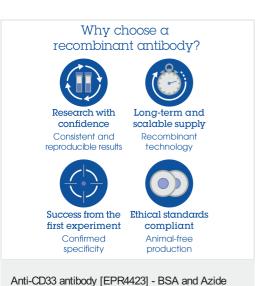
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (<u>ab134115</u>).



Flow Cytometry - Anti-CD33 antibody [EPR4423] - BSA and Azide free (ab214439)



Immunoprecipitation - Anti-CD33 antibody
[EPR4423] - BSA and Azide free (ab214439)



free (ab214439)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

#### Our Abpromise to you: Quality guaranteed and expert technical support

- · Replacement or refund for products not performing as stated on the datasheet
- · Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- · We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit <a href="https://www.abcam.com/abpromise">https://www.abcam.com/abpromise</a> or contact our technical team.

#### Terms and conditions

• Guarantee only valid for products bought direct from Abcam or one of our authorized distributors