abcam

ab214439

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

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR4423
Purification technique	Affinity purification Protein A
Dissociation constant	1.76×10^{-11} M
Concentration	1.042 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	-

Notes	-

Flow Cyt

Tested

Species	Human
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

IHC-P

Not recommended

Species	Human
Dilution info	-
Notes	-

Target data

See full target information CD33

Function	Sialic-acid-binding immunoglobulin-like lectin (Siglec) that plays a role in mediating cell-cell interactions and in maintaining immune cells in a resting state (PubMed:10611343, PubMed:11320212, PubMed:15597323). Preferentially recognizes and binds alpha-2,3- and more avidly alpha-2,6-linked sialic acid-bearing glycans (PubMed:7718872). Upon engagement of ligands such as C1q or syalylated glycoproteins, two immunoreceptor tyrosine-based inhibitory motifs (ITIMs) located in CD33 cytoplasmic tail are phosphorylated by Src-like kinases such as LCK (PubMed:10887109, PubMed:28325905). These phosphorylations provide docking sites for the recruitment and activation of protein-tyrosine phosphatases PTPN6/SHP-1 and PTPN11/SHP-2 (PubMed:10206955, PubMed:10556798, PubMed:10887109). In turn, these phosphatases regulate

downstream pathways through dephosphorylation of signaling molecules (PubMed:10206955, PubMed:10887109). One of the repressive effect of CD33 on monocyte activation requires phosphoinositide 3-kinase/PI3K (PubMed:15597323).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab214439 is the carrier-free version of ab134115.

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

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. 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.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

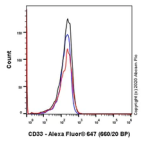
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

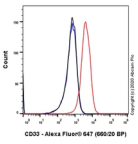
In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
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3 product images



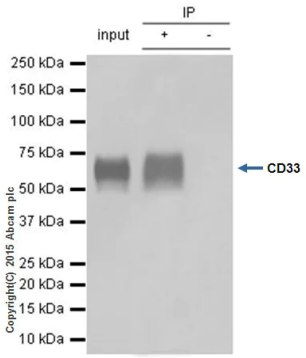


Flow Cytometry - Anti-CD33 antibody [EPR4423] - BSA and Azide free (ab214439)

CD33 Flow Cytometry staining using rabbit Anti-CD33 antibody

Flow Cytometry analysis of Jurkat (Human T cell leukemia T lymphocyte, Left) / THP1 (Human monocytic leukemia monocyte, Right) labeling CD33 with ab134115 at 1/500 dilution (red). Goat anti rabbit IgG (Alexa Fluor® 647, ab150079) at 1/2000 dilution was used as the secondary antibody. Rabbit monoclonal IgG (ab172730) 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).

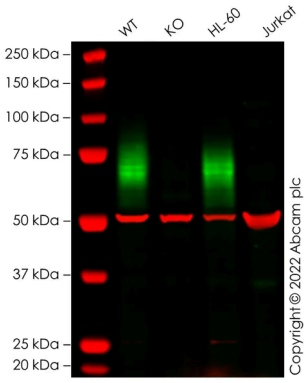


Immunoprecipitation - Anti-CD33 antibody [EPR4423] - BSA and Azide free (ab214439)

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

All lanes:
Immunoprecipitation - Anti-CD33 antibody [EPR4423] (ab134115)

Predicted band size: 40 kDa



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

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

Lane 1:
Wild-type THP-1 cell lysate at 10 µg

Lane 2:
Western blot - Human CD33 knockout THP-1 cell lysate (ab273785) at 10 µg

Lane 3:
HL-60 cell lysate at 10 µg

Lane 4:
Jurkat cell lysate at 10 µg

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

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.