abcam

ab221678

Anti-CD14 antibody [EPR21847]

Anti-CD14 antibody [EPR21847] (ab221678) is a rabbit monoclonal antibody detecting CD14 in **Western Blot, Flow Cytometry, IP, IHC-Fr, ICC/IF**. Suitable for **Mouse**.

- Biophysical QC for unrivalled batch-batch consistency
- Over 10 publications

Recombinant | RabMAb | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR21847
Purification technique	Affinity purification Protein A
Concentration	0.517 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Mouse

Dilution info	1/30
Notes	-

Flow Cyt

Tested

Species	Mouse
Dilution info	1/500
Notes	-

WB

Tested

Species	Mouse
Dilution info	1/1000
Notes	-

ICC/IF

Tested

Species	Mouse
Dilution info	1/1000
Notes	-

IHC-Fr

Tested

Species	Mouse
Dilution info	-
Notes	-

Target data

See full target information Cd14

Function	Coreceptor for bacterial lipopolysaccharide. In concert with LBP, binds to monomeric lipopolysaccharide and delivers it to the LY96/TLR4 complex, thereby mediating the innate immune response to bacterial lipopolysaccharide (LPS) (PubMed:16148141). Acts via MyD88, TIRAP and TRAF6, leading to NF-kappa-B activation, cytokine secretion and the inflammatory response (PubMed:15895089, PubMed:8612135). Acts as a coreceptor for TLR2:TLR6 heterodimer in response to diacylated lipopeptides and for TLR2:TLR1 heterodimer in response to triacylated lipopeptides, these clusters trigger signaling from the cell surface and subsequently are targeted to the Golgi in a lipid-raft dependent pathway (By similarity). Acts as an accessory receptor for M.tuberculosis lipoproteins LprA, LprG and LpqH, in conjunction with coreceptors TLR2 and TLR1. The lipoproteins act as agonists to modulate antigen presenting cell functions in response to the pathogen (PubMed:19362712). Binds electronegative LDL (LDL(-)) and mediates the cytokine release induced by LDL(-) (By similarity).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-CD14 antibody [EPR21847] (ab221678) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Flow Cyt), Immunoprecipitation (IP), Immunohistochemistry (IHC-Fr), Immunocytochemistry/immunofluorescence (ICC/IF) in Mouse samples.

What is the molecular weight of CD14?
Anti-CD14 [EPR21847] (ab221678) specifically detects a band for CD14 (UniProt: P10810) at a molecular weight of 39kDa.

Trusted by the scientific community
Anti-CD14 [EPR21847] (ab221678) was first used in a scientific publication in 2018 and has been cited over 10 times in peer-reviewed journals.

Other related products
We have a range of other formats of antibody clone [EPR21847] also available for your convenience: ab221678, Carrier free - ab231852, Alexa Fluor® 488 - ab237391, Alexa Fluor® 647 - ab302829, Alexa Fluor® 594 - ab305117

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.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	CD14 also known as cluster of differentiation 14 is a glycoprotein with a molecular mass of approximately 55 kDa. This protein is expressed mainly on the surface of macrophages monocytes and to a lesser extent on neutrophils. CD14 exists in two forms: membrane-bound (mCD14) and soluble (sCD14). It functions as a co-receptor along with TLR4 (Toll-like receptor 4) and MD-2 for the detection of bacterial lipopolysaccharide (LPS) which is an important component of the outer membrane of Gram-negative bacteria.
Biological function summary	This glycoprotein plays a significant role in the innate immune system by recognizing pathogen-associated molecular patterns (PAMPs). It is part of a receptor complex where CD14 acts alongside LPS binding protein (LPS-R) TLR4 and MD-2 to facilitate pro-inflammatory signaling in response to microbial invasion. By binding to LPS CD14 initiates and amplifies the immune response leading to the production of cytokines and the recruitment of other immune cells to the site of infection.
Pathways	CD14 interacts with the toll-like receptor signaling pathway notably involving TLR4. Upon LPS detection CD14 assists in the activation of TLR4 triggering downstream signaling cascades such as NF-κB and MAPK pathways. These pathways result in the transcription of inflammatory cytokines and are essential for the host defense mechanism. The protein complex involving CD14 further interacts with adaptor proteins like MyD88 and TRIF facilitating signal propagation and the immune system's ability to respond to pathogens.
Associated diseases and disorders	Researchers have linked CD14 to sepsis and atherosclerosis. High levels of soluble CD14 (sCD14) are often observed in patients with sepsis indicating its participation

in excessive systemic inflammation. In atherosclerosis CD14's role in recognizing LPS contributes to chronic inflammation and the development of plaques in arteries. Through these diseases CD14 connects with TLR4 emphasizing its impact on inflammatory responses and its potential as a therapeutic target.

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

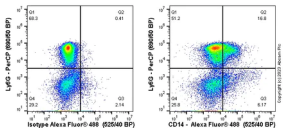
We do not recommend this combination. It is not covered by our product promise.

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:
Terms & Conditions.

8 product images



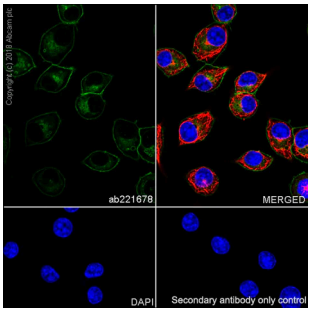
Flow Cytometry - Anti-CD14 antibody [EPR21847] (ab221678)

CD14 Flow Cytometry staining using rabbit Anti-CD14 antibody

Flow cytometry staining of C57 BL/6 mouse bone marrow cells with ab221678 (right) or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (left). Cells were incubated for 30 min on ice in 1x PBS containing 10µg/ml anti CD16/CD32 and 10% normal goat serum to block FC receptors and non-specific protein-protein interaction followed by the antibody ab221678 or Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control (1x 106 in 100 µl at 10.0 µg/ml (1/215)) for 30min on ice. The cells were simultaneously stained with Ly6G.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min on ice

Acquisition of >30000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter. Events were gated on viable cells.

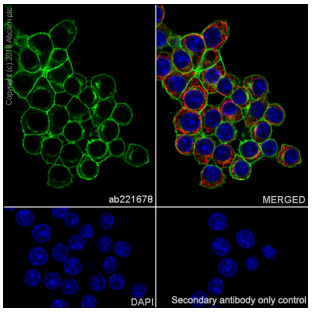


Immunocytochemistry/ Immunofluorescence - Anti-CD14 antibody [EPR21847] (ab221678)

CD14 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CD14 antibody

Immunofluorescent analysis of 100% methanol-fixed J774A.1 (mouse reticulum cell sarcoma monocyte macrophage) cells labeling CD14 with ab221678 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing membranous and cytoplasmic staining on J774A.1 cell line. The nuclear counter stain is DAPI (blue).
Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary at 1/1000 dilution.

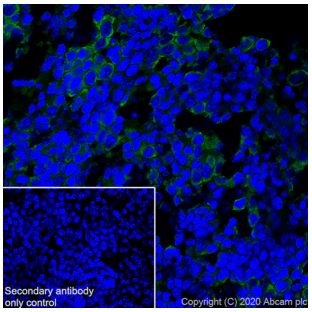


Immunocytochemistry/ Immunofluorescence - Anti-CD14 antibody [EPR21847] (ab221678)

CD14 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CD14 antibody

Immunofluorescent analysis of 100% methanol-fixed RAW 264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) cells labeling CD14 with ab221678 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488)(ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing membranous and cytoplasmic staining on RAW 264.7 cell line. The nuclear counter stain is DAPI (blue).
Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) at 1/200 dilution (re

Secondary antibody only control: Used PBS instead of primary antibody, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488)(ab150077) secondary at 1/1000 dilution.

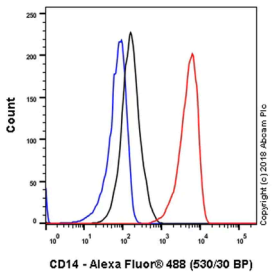


Immunohistochemistry (Frozen sections) - Anti-CD14 antibody [EPR21847] (ab221678)

CD14 Immunohistochemistry (Frozen sections) staining using rabbit Anti-CD14 antibody

Immunohistochemical analysis of 4% PFA-fixed, 0.2% Triton X-100 permeabilized frozen Mouse spleen tissue labeling CD14 with ab221678 at 1/50 (10.62 ug/ml) dilution followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor ® 488) at 1/1000 dilution (Green). Positive staining on mouse spleen. is observed. The nuclear counterstain was DAPI (Blue).
Secondary antibody control: Secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor ®; 488) at 1000 dilution.

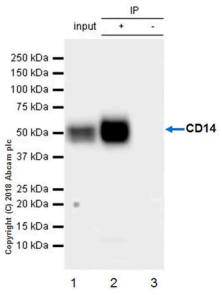
Heat mediated antigen retrieval using sodium citrate buffer (10mM citrate pH 6.0 + 0.05% Tween-20).



Flow Cytometry - Anti-CD14 antibody [EPR21847] (ab221678)

CD14 Flow Cytometry staining using rabbit Anti-CD14 antibody

Flow cytometric analysis of RAW 264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) cell line labeling CD14 with ab221678 at 1/500 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabelled control (cells incubated with secondary antibody only) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.
Gated on viable cells.



Immunoprecipitation - Anti-CD14 antibody [EPR21847] (ab221678)

CD14 was immunoprecipitated from 0.35 mg RAW 264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate with ab221678 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab221678 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/5000 dilution.
Lane 1: RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate 10 µg (Input).
Lane 2: ab221678 IP in RAW 264.7 whole cell lysate(+).
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab221678 in RAW 264.7 whole cell lysate (-).

Blocking and dilution buffer and concentration: 5% NFDM/TBST.
Exposure time: 8 seconds

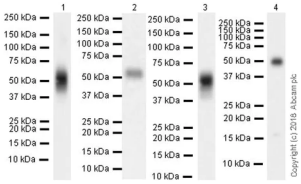
The molecular mass observed is consistent with the literature (PMID: 9502426; PMID:7513013)

All lanes:
Immunoprecipitation - Anti-CD14 antibody [EPR21847] (ab221678)

Developed using the ECL technique.

Predicted band size: 40 kDa

Observed band size: 50-55 kDa



Western blot - Anti-CD14 antibody [EPR21847] (ab221678)

Blocking/Dilution buffer and concentration: 5% NFDM/TBST.
Exposure time: Lane 1: 6 seconds; Lane 2: 3 minutes; Lane 3: 10 seconds; Lane 4: 81 seconds.

The molecular mass observed is consistent with the literature (PMID: 9502426; PMID: 7513013).

All lanes:
Western blot - Anti-CD14 antibody [EPR21847] (ab221678) at 1/1000 dilution

Lane 1:
J774A.1 (mouse reticulum cell sarcoma monocyte macrophage), whole cell lysate at 10 µg

Lane 2:
Mouse lymph node lysate at 20 µg

Lane 3:
RAW264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage), whole cell lysate at 10 µg

Lane 4:

Mouse placenta lysate at 10 µg

Secondary

Lanes 1 - 3:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Lane 4:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/50000 dilution

Developed using the ECL technique.

Predicted band size: 40 kDa

Observed band size: 50-55 kDa

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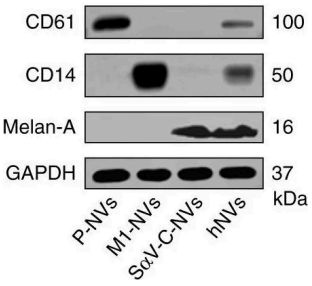


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Western blot - Anti-CD14 antibody [EPR21847] (ab221678)

CD14 Western blot staining using rabbit Anti-CD14 antibody

CD14 western blot using anti-CD14 antibody [EPR21847] ab221678. Publication image and figure legend from Rao, L., Wu, L., et al., 2020, Nat Commun, PubMed 32999291.

ab221678 was used in this publication in western blot. This may not be the same as the application(s) guaranteed by Abcam. For a full list of applications guaranteed by Abcam for ab221678 please see the product overview.

Schematic and characterization of hNVs.a Schematic showing the hNVs consist of engineered SαV-C-NVs, M1-NVs, and P-NVs. b Schematic showing the hNVs efficiently interact with CTCs in the blood, accumulate in the post-surgical tumor bed, repolarize TAMs towards M1 phenotype, and block the CD47-SIRPα 'don't eat me' pathway, thus promoting macrophage phagocytosis of cancer cells, as well as boosting antitumor Tcell immunity. c, d Immunofluorescence imaging c and flow cytometry analysis d of SαV expression on original and engineered B16F10 cells. Scale bar, 10 µm. e Relative mRNA expression of Cd86, Tnf, Il6, and Inos in M0 cells, M1 cells, M0-NVs and M1-NVs. f Hydrodynamic size and zeta potential of P-NVs, M1-NVs, SαV-C-NVs, and hNVs measured by DLS. g TEM images of hNVs. Scale bar, 100 nm and 50 nm in the left and right panel, respectively. The samples were negatively stained with uranyl acetate. h Immunofluorescence images of hNVs. Scale bar, 10 µm. i Western blot analysis of specific protein CD61, CD14, and Melan-A in the samples of P-NVs, M1-NVs, SαV-C-NVs, and hNVs. All data are presented as mean ± S.D. (n = 3). Statistical significance was calculated via unpaired two-tailed t test. **p < 0.01; ***p < 0.001.

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