# abcam

# Product datasheet

# Anti-CD14 antibody [EPR3653] ab133335



★★★★★ 1 Abreviews 22 References 11 Images

#### Overview

**Product name** Anti-CD14 antibody [EPR3653]

**Description** Rabbit monoclonal [EPR3653] to CD14

**Host species** Rabbit

**Tested applications** Suitable for: WB. IHC-P

Unsuitable for: ICC/IF

Species reactivity Reacts with: Human

**Immunogen** Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control IHC-P: human colon, placenta and tonsil tissue; WB: Human tonsil tissue lysate, SW480 and

PBMCI 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  $\mathsf{RabMAb}^{\texttt{®}}$  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.

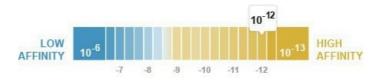
### **Properties**

**Form** Liquid

Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long Storage instructions

term. Avoid freeze / thaw cycle.

 $K_D = 4.70 \times 10^{-12} M$ Dissociation constant (K<sub>D</sub>)



## Learn more about K<sub>D</sub>

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

ClonalityMonoclonalClone numberEPR3653

**Isotype** IgG

## **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab133335 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		1/1000 - 1/5000. Detects a band of approximately 53 kDa (predicted molecular weight: 40 kDa).
IHC-P	<b>★★★★☆ (1)</b>	1/2000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.  See IHC antigen retrieval protocols.  For unpurified use at 1/500 - 1/1000.

**Application notes** Is unsuitable for ICC/IF.

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**Function** Cooperates with MD-2 and TLR4 to mediate the innate immune response to bacterial

lipopolysaccharide (LPS). Acts via MyD88, TIRAP and TRAF6, leading to NF-kappa-B activation, cytokine secretion and the inflammatory response. Up-regulates cell surface molecules, including

adhesion molecules.

**Tissue specificity** Expressed strongly on the surface of monocytes and weakly on the surface of granulocytes; also

expressed by most tissue macrophages.

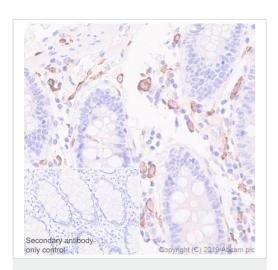
Sequence similarities Contains 11 LRR (leucine-rich) repeats.

Post-translational modifications

N- and O- glycosylated. O-glycosylated with a core 1 or possibly core 8 glycan.

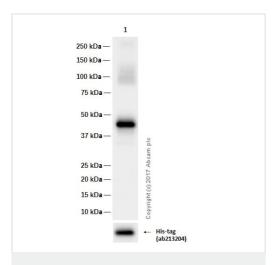
**Cellular localization** Cell membrane.

# Images



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD14 antibody
[EPR3653] (ab133335)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue sections labeling CD14 with purified ab133335 at 1/2000 dilution (0.04 µg/ml). Heat mediated antigen retrieval was performed using <a href="mailto:ab93684">ab93684</a> (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Western blot - Anti-CD14 antibody [EPR3653] (ab133335)

Anti-CD14 antibody [EPR3653] (ab133335) at 1/1000 dilution (purified) + His-Tagged Human CD14 (aa20 to 345) recombinant protein at  $0.015~\mu g$ 

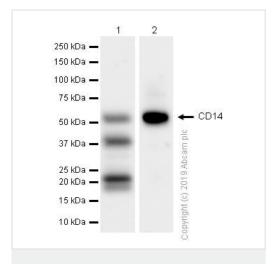
# **Secondary**

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

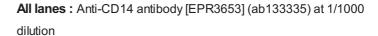
Predicted band size: 40 kDa Observed band size: 43 kDa

Exposure time: 1 second

Blocking and diluting buffer: 5% NFDM/TBST



Western blot - Anti-CD14 antibody [EPR3653] (ab133335)



Lane 1: Human tonsil lysates prepared in RIPA lysis method

Lane 2: Human tonsil lysates prepared in 1%SDS Hot lysis

method

Lysates/proteins at 20 µg per lane.

# **Secondary**

 $\begin{tabular}{ll} \textbf{All lanes:} Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution \end{tabular}$ 

Predicted band size: 40 kDa

Exposure time: 45 seconds

Blocking/Diluting buffer and concentration: 5% NFDM/TBST

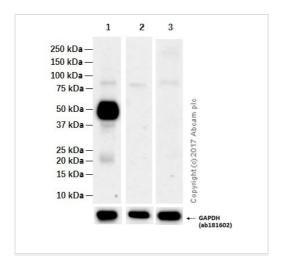
**All lanes :** Anti-CD14 antibody [EPR3653] (ab133335) at 1/1000 dilution (purified)

**Lane 1**: Human tonsil tissue lysate prepared in 1% SDS Hot lysis method

Lane 2 : HeLa (Human cervix adenocarcinoma) whole cell lysate

Lane 3: U-937 (Human histiocytic lymphoma) whole cell lysate

Lysates/proteins at 20 µg per lane.



Western blot - Anti-CD14 antibody [EPR3653] (ab133335)

## Secondary

**Lane 1**: Goat Anti-Rabbit lgG (HRP) with minimal cross-reactivity with human lgG at 1/2000 dilution

Lanes 2-3: Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at

#### 1/20000 dilution

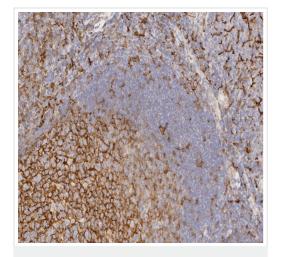
**Predicted band size:** 40 kDa **Observed band size:** 53 kDa

Exposure time: 3 minutes

Blocking and diluting buffer: 5% NFDM/TBST

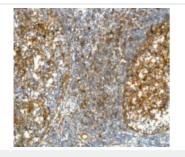
The expression level in HeLa and U-937 are low (PMID: 9886426

and 15730927)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD14 antibody
[EPR3653] (ab133335)

Immunohistochemical analysis of Formalin-fixed, paraffinembedded human tonsil tissue labelling CD14 with ab133335 (unpurified) at 1/500 dilution. No blocking step performed. Anti-Rabbit HRP polymer was used as the secondary detection system. Heat-mediated antigen retrieval was performed using EDTA based pH 9.0 buffer.



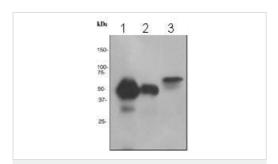
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD14 antibody
[EPR3653] (ab133335)

Immunohistochemical analysis of Formalin-fixed, paraffinembedded Human tonsil tissue labelling CD14 with ab133335 (unpurified) at 1/500 dilution. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD14 antibody
[EPR3653] (ab133335)

Immunohistochemical analysis of Formalin-fixed, paraffinembedded Human placenta tissue labelling CD14 with ab133335 (unpurified) at 1/500 dilution. Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



Western blot - Anti-CD14 antibody [EPR3653] (ab133335)

**All lanes :** Anti-CD14 antibody [EPR3653] (ab133335) at 1/1000 dilution (unpurified)

Lane 1: PBMC cell lysate prepared in 1% SDS Hot lysis method
Lane 2: Human tonsil tissue lysate prepared in 1%SDS Hot lysis
method

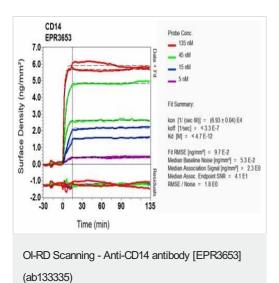
Lane 3: SW480 cell lysate prepared in 1%SDS Hot lysis method

Lysates/proteins at 10 µg per lane.

# **Secondary**

All lanes: Goat Anti-rabbit HRP at 1/2000 dilution

**Predicted band size:** 40 kDa **Observed band size:** 53 kDa



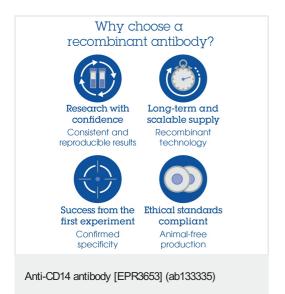
Equilibrium disassociation constant  $(K_D)$ Learn more about  $K_D$ 

# Click here to learn more about K<sub>D</sub>

Normal fissue samples				Malignant tissue samples			
Human cardiac muscle	x	Human placenta	x (immune cels √)	Clear cell carcinoma of human kidney	x [immune cels √]	Human glioma	✓
Human cerebrum	x	Human skeletal muscle	x (immune cels √)	Human bladder cancer		Human hepatocellular carcinoma	× (immune cells √)
Human colon	x (immune cells √)	Human skin	x (immune cells √)	Human breast carcinoma	x [mmune cels √]	Human lung carcinoma	x (immune cells √)
Human endometrium	× (immune cells √)	Human spleen	× (immune cells ✓)	Human cervical carcinoma	¥ [immune cells √]	Human ovarian carcinoma	× (immune cells √)
Human kidney	× (immune cells √)	Human stomach	× (immune cells ✓)	Human colon carcinoma	x [immune cels √]	Human pancreatic carcinoma	x (immune cells √)
Human liver	x (immune cells √)	Human testis	x (immune cels √)	Human endometrial carcinoma	▼ [immune cels √]	Human prostatic hyperplasia	x (immune cells √)
Human lung	x (immune cells √)	Human thyroid	×	Human gastric adenocarcinoma	▼ [immune cells √]	Human thyroid carcinoma	×
luman mammary gland	× (mmune cells √)	Human tonsi	x (immune cells √)				
Human pancreas	x (immune cells √)						

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CD14 antibody
[EPR3653] (ab133335)

Tissue Microarrays stained for "Anti-CD14 antibody [EPR3653]" using "ab133335" in immunohistochemical analysis. This table provides a detailed overview of positive (tick mark) and negative (cross mark) staining per sample type tested. The sections were pre-treated using Heat mediated antigen retrieval using Bond™ Epitope Retrieval Solution 2 (pH 9.0) for 20 minutes. The sections were incubated with ab133335 for 30 mins at room temperature followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



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