# abcam

## Product datasheet

## Anti-TLR9 antibody [EPR14964-2] ab187148



Recombinant RabMAb

## 5 Images

#### Overview

**Product name** Anti-TLR9 antibody [EPR14964-2]

**Description** Rabbit monoclonal [EPR14964-2] to TLR9

**Host species** Rabbit

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

Unsuitable for: Flow Cyt (Intra) or ICC/IF

Species reactivity Reacts with: Human

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Daudi, Raji and Ramos cell lysates. IHC-P: Human tonsil and breast carcinoma tissues.

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

## **Properties**

**Form** Liquid

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

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

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

**Purity** Protein A purified

Clonality Monoclonal EPR14964-2 Clone number

Isotype ΙgG

## **Applications**

## The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab187148 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 130 kDa (predicted molecular weight: 116 kDa).
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

## **Application notes**

Is unsuitable for Flow Cyt (Intra) or ICC/IF.

### **Target**

Function Key component of innate and adaptive immunity. TLRs (Toll-like receptors) control host immune

response against pathogens through recognition of molecular patterns specific of

microorganisms. TLR9 is a nucleotide-sensing TLR which is activated by unmethylated cytidine-phosphate-guanosine (CpG) dinucleotides. Acts via MYD88 and TRAF6, leading to NF-kappa-B

activation, cytokine secretion and the inflammatory response.

Tissue specificity Highly expressed in spleen, lymph node, tonsil and peripheral blood leukocytes, especially in

plasmacytoid pre-dendritic cells. Levels are much lower in monocytes and CD11c+ immature

dendritic cells. Also detected in lung and liver.

**Sequence similarities**Belongs to the Toll-like receptor family.

Contains 26 LRR (leucine-rich) repeats.

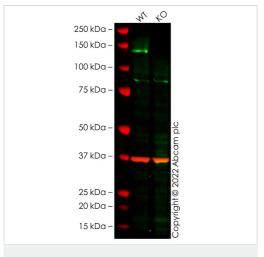
Contains 1 TIR domain.

**Cellular localization** Endoplasmic reticulum membrane. Endosome. Lysosome. Cytoplasmic vesicle > phagosome.

Relocalizes from endoplasmic reticulum to endosome and lysosome upon stimulation with

agonist.

### **Images**



Western blot - Anti-TLR9 antibody [EPR14964-2] (ab187148)

**All lanes :** Anti-TLR9 antibody [EPR14964-2] (ab187148) at 1/1000 dilution

Lane 1: Wild-type Raji cell lysate

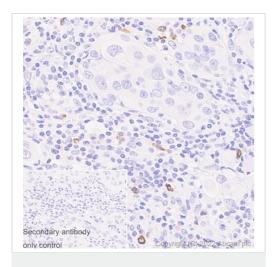
Lane 2: TLR9 knockout Raji cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 116 kDa **Observed band size:** 140 kDa

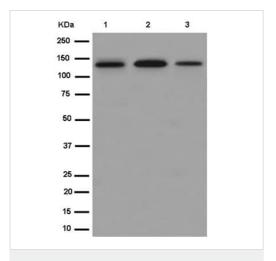
False colour image of Western blot: Anti-TLR9 antibody [EPR14964-2] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab187148 was shown to bind specifically to TLR9. A band was observed at 140 kDa in wild-type Raji cell lysates with no signal observed at this size in TLR9 knockout cell line ab280879 (knockout cell lysate ab282939). To generate this image, wild-type and TLR9 knockout Raji cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in fluorescent western blot (TBS-based) blocking solution 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.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TLR9 antibody
[EPR14964-2] (ab187148)

Immunohistochemistry analysis of paraffin-embedded Human breast carcinoma tissue sections labelling TLR9 with ab187148 at 1/1000 dilution. The section was incubated with ab187148 for 30 mins at room temperature. Ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used as the secondary antibody. Sections were counterstained with Hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

Positive staining on some immune stroma cells in human breast carcinoma tissue. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



Western blot - Anti-TLR9 antibody [EPR14964-2] (ab187148)

**All lanes :** Anti-TLR9 antibody [EPR14964-2] (ab187148) at 1/5000 dilution

Lane 1 : Daudi cell lysate
Lane 2 : Raji cell lysate
Lane 3 : Ramos cell lysate

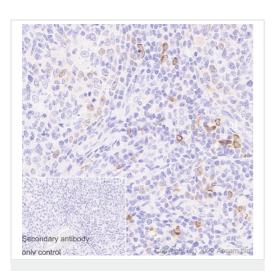
Lysates/proteins at 10 µg per lane.

## Secondary

**All lanes :** Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 116 kDa

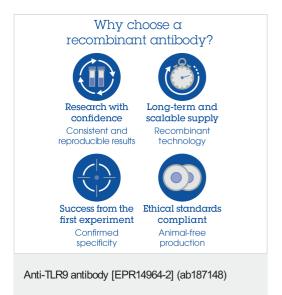
Blocking and dilution buffer: 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TLR9 antibody
[EPR14964-2] (ab187148)

Immunohistochemistry analysis of paraffin-embedded Human tonsil tissue sections labelling TLR9 with ab187148 at 1/1000 dilution. The section was incubated with ab187148 for 30 mins at room temperature. Ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used as the secondary antibody. Sections were counterstained with Hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins.

Positive staining on some immune cells in human tonsil tissue. The immunostaining was performed on a Leica Biosystems BOND® RX instrument.



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