abcam

Product datasheet

Anti-IRAK-1 antibody [EPR18630] ab180747



Recombinant RabMAb

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Overview

Product name Anti-IRAK-1 antibody [EPR18630]

Description Rabbit monoclonal [EPR18630] to IRAK-1

Host species Rabbit

Tested applications Suitable for: WB, IP

Unsuitable for: ICC/IF

Reacts with: Human Species reactivity

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

Positive control WB: HEK-293, Jurkat, K562 and HeLa whole cell lysates. ICC/IF: HeLa and K562 cells. IP: HEK-

293 whole cell lysate.

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 Preservative: 0.01% Sodium azide

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

Purity Protein A purified

Clonality Monoclonal Clone number EPR18630

Isotype ΙgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab180747 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. Detects a band of approximately 77 kDa (predicted molecular weight: 77 kDa).	
IP		1/100.	

Application notes

Is unsuitable for ICC/IF.

Target

	on

Binds to the IL-1 type I receptor following IL-1 engagement, triggering intracellular signaling cascades leading to transcriptional up-regulation and mRNA stabilization. Isoform 1 binds rapidly but is then degraded allowing isoform 2 to mediate a slower, more sustained response to the cytokine. Isoform 2 is inactive suggesting that the kinase activity of this enzyme is not required for IL-1 signaling. Once phosphorylated, IRAK1 recruits the adapter protein PELI1.

Tissue specificity

lsoform 1 and isoform 2 are ubiquitously expressed in all tissues examined, with isoform 1 being more strongly expressed than isoform 2.

Sequence similarities

Belongs to the protein kinase superfamily. TKL Ser/Thr protein kinase family. Pelle subfamily.

Contains 1 protein kinase domain.

Post-translational modifications

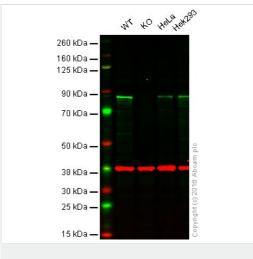
Autophosphorylated or is transphosphorylated by IRAK4 following recruitment to the IL-1RI. In the

case of isoform 1, this is linked to ubiquitination and degradation.

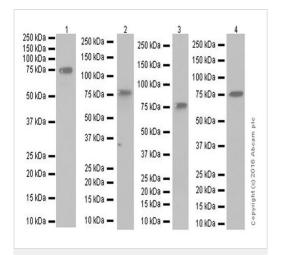
Polyubiquitinated; after cell stimulation with IL-1-beta. Polyubiquitination occurs with polyubiquitin

chains linked through 'Lys-63'.

Images



Western blot - Anti-IRAK-1 antibody [EPR18630] (ab180747)



Western blot - Anti-IRAK-1 antibody [EPR18630] (ab180747)

Lane 1: Wild type HAP1 whole cell lysate (20 µg)

Lane 2: IRAK-1 knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: Hek293 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab180747 observed at 85 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab180747 was shown to specifically react with IRAK-1 when IRAK-1 knockout samples were used. Wild-type and IRAK-1 knockout samples were subjected to SDS-PAGE. Ab180747 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10000 dilution respectively. Blots were developed with 800CW Goat anti-Rabbit and 680CW Goat anti-Mouse secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.

All lanes : Anti-IRAK-1 antibody [EPR18630] (ab180747) at 1/1000 dilution

Lane 1 : HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lane 2 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lane 3 : K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate

Lane 4 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 77 kDa **Observed band size:** 77 kDa

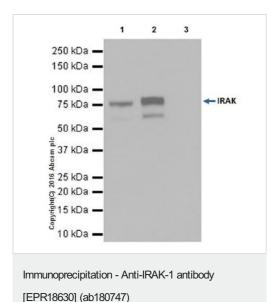
Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: Lane 1: 10 seconds; Lane 2: 3 minutes; Lane 3: 30

seconds; Lane 4: 1 minute.

There are two splice variants for IRAK-1, IRAK-1b and IRAK-1c,

respectively. The product recognizes full length IRAK-1 (80kDa).



IRAK-1 was immunoprecipitated from 0.35 mg of HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate with ab180747 at 1/100 dilution. Western blot was performed from the immunoprecipitate using ab180747 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1000 dilution.

Lane 1: HEK-293 whole cell lysate 10µg (Input).

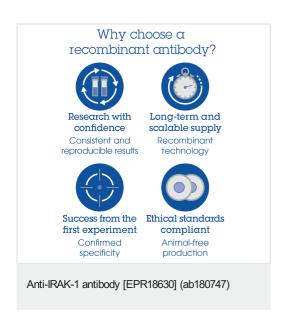
Lane 2: ab180747 IP in HEK-293 whole cell lysate.

Lane 3: Rabbit monoclonal $\lg G$ ($\underline{ab172730}$) instead of ab180747 in HEK-293 whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds.

The 68kDa band observed is likely a splice variant, according to PMID: 16690127.



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