abcam

ab119942

Anti-IRAK4 antibody [2H9]

Anti-IRAK4 antibody [2H9] (ab119942) is a mouse monoclonal antibody detecting IRAK4 in **Western Blot, Flow Cytometry, IHC-P, ICC/IF, ELISA**. Suitable for **African green monkey, Human, Mouse,**.

- KO validated for confirmed specificity

KO Validated

Key facts

Isotype	IgG1
Host species	Mouse
Storage buffer	Preservative: 0.05% Sodium azide Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	Recombinant Fragment Protein within Human IRAK4 aa 1 to C-terminus. The exact immunogen used to generate this antibody is proprietary information. Database link Q9NWZ3
Clone number	2H9
Purification technique	Affinity purification Protein G
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →
Purification notes	Purified from tissue culture supernatant.

Reactivity data

Flow Cyt

Tested

Species	Human
Dilution info	1/200 - 1/400
Notes	ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	-
Notes	-

Species	African green monkey
Dilution info	-
Notes	ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Not recommended

Species	Recombinant fragment - Human, Recombinant full length protein - Human
Dilution info	-
Notes	-

ELISA

Tested

Species	Recombinant fragment - Human
Dilution info	1/10000
Notes	-

Expected

Species	Human, Mouse, African green monkey
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant full length protein - Human
Dilution info	-
Notes	-

WB

Tested

Species	Recombinant full length protein - Human
Dilution info	1/500 - 1/2000
Notes	-

Species	Mouse
Dilution info	1/500 - 1/2000
Notes	-

Species	African green monkey
Dilution info	1/500 - 1/2000
Notes	-

Species	Human
Dilution info	1/500 - 1/2000
Notes	-

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human

Dilution info	1/200 - 1/1000
Notes	-

Expected

Species	Mouse, African green monkey
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human, Recombinant full length protein - Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100
Notes	-

Expected

Species	Mouse, African green monkey
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human, Recombinant full length protein - Human
Dilution info	-
Notes	-

Target data

See full target information IRAK4

Function	Serine/threonine-protein kinase that plays a critical role in initiating innate immune response against foreign pathogens. Involved in Toll-like receptor (TLR) and IL-1R signaling pathways (PubMed:17878374). Is rapidly recruited by MYD88 to the receptor-signaling complex upon TLR activation to form the Myddosome together with IRAK2. Phosphorylates initially IRAK1, thus stimulating the kinase activity and intensive autophosphorylation of IRAK1. Phosphorylates E3 ubiquitin ligases Pellino proteins (PELI1, PELI2 and PELI3) to promote pellino-mediated polyubiquitination of IRAK1. Then, the ubiquitin-binding domain of IKBKG/NEMO binds to polyubiquitinated IRAK1 bringing together the IRAK1-MAP3K7/TAK1-TRAF6 complex and the NEMO-IKKA-IKKB complex. In turn, MAP3K7/TAK1 activates IKKs (CHUK/IKKA and IKBKB/IKKB) leading to NF-kappa-B nuclear translocation and activation. Alternatively, phosphorylates TIRAP to promote its ubiquitination and subsequent degradation. Phosphorylates NCF1 and regulates NADPH oxidase activation after LPS stimulation suggesting a similar mechanism during microbial infections.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C

Notes

What is this antibody validated in?
Anti-IRAK4 antibody [2H9] (ab119942) is a mouse monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Flow Cyt), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF), ELISA in African green monkey, Human, Mouse, samples.

What is the molecular weight of IRAK4?
Anti-IRAK4 [2H9] (ab119942) specifically detects a band for IRAK4 (UniProt: Q9NWZ3) at a molecular weight of 52kDa.

Specificity confirmed
The specificity of Anti-IRAK4 antibody [2H9] (ab119942) has been confirmed by Western blot testing in IRAK4 Knockout THP-1 cell line, ab281630.

Supplementary info

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

Activity summary	IRAK4 also known as Interleukin-1 receptor-associated kinase 4 is an important protein with an approximate molecular mass of 51 kDa. This protein is a significant part of the signaling cascade initiated by the Interleukin-1 receptor (IL-1R) and Toll-like receptors (TLRs). Expressed in a variety of tissues including the spleen thymus and lung IRAK4 is involved in innate immune responses. The presence of IRAK4 protein facilitates the downstream signaling pathways that activate the transcription factors responsible for inflammatory cytokine production.
Biological function summary	IRAK4 plays an important role in mediating signaling in the innate immune system by forming complexes with other proteins such as IRAK-1. These complexes propagate signals that result in the activation of transcription factors like NF-kB and AP-1 which regulate the expression of inflammatory genes. Through these mechanisms IRAK4 influences the body's ability to respond to infections and stresses. Its critical involvement in these signaling pathways highlights its importance in maintaining immune homeostasis.
Pathways	IRAK4 is a pivotal player in the Toll-like receptor and IL-1 receptor signaling pathways which are central to the host defense against pathogens. It functions upstream activating the MyD88-dependent pathway resulting in the recruitment and phosphorylation of other kinases such as IRAK-1 and TRAF6. These interactions trigger the NF-kB signaling cascade thereby enhancing pro-inflammatory cytokine production that is essential for initiating immune responses.
Associated diseases and disorders	Mutations or dysregulation of IRAK4 are linked to increased susceptibility to recurrent bacterial infections and sepsis. Patients with IRAK4 deficiency have impaired NF-kB activation leading to a weakened inflammatory response. Additionally overexpression of IRAK4 has been observed in certain autoimmune diseases such as rheumatoid arthritis where the persistent activation of TLR signaling contributes to chronic inflammation. The role of IRAK4 in these disorders ties closely with its interaction with proteins like TRAF6 influencing disease development and progression.

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.

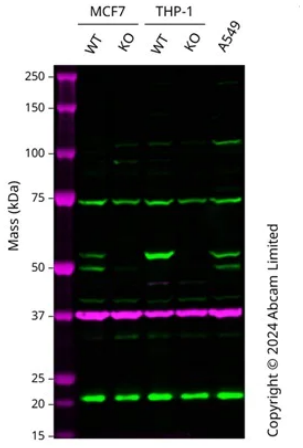
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

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9 product images



Western blot - Anti-IRAK4 antibody [2H9] (ab119942)

IRAK4 Western blot staining using mouse Anti-IRAK4 antibody

Lane 1: Wild-type MCF7 cell lysate 20 ug Lane 2: IRAK4 knockout MCF7 cell lysate 20 ug Lane 3: Wild-type THP-1 cell lysate 20 ug Lane 4: IRAK4 knockout THP-1 cell lysate 20 ug Lane 5: A549 cell lysate 20 ug Western blot: Anti-IRAK4 antibody [2H9] (ab119942) staining at 1/1000 dilution, shown in green; Rabbit Anti-GAPDH antibody [EPR16891] (ab181602) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab119942 was shown to bind specifically to IRAK4. A band was observed at 52 kDa in wild-type MCF7 cell lysates and wild-type THP-1 cell lysates with no signal observed at this size in IRAK4 knockout cell line. To generate this image, wild-type and IRAK4 knockout MCF7 cell lysates and knockoutTHP-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-Mouse IgG H&L 800CW and Goat anti-Rabbit IgG H&L 680RD at 1/20000 dilution.
All lanes:
Western blot - Anti-IRAK4 antibody [2H9] (ab119942) at 1/1000 dilution

Lane 1:
Wild-type MCF7 cell lysate at 20 µg

Lanes 2 and 4:
Western blot - Human IRAK4 knockout THP-1 cell line (ab281630) at 20 µg

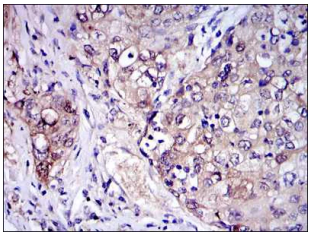
Lane 5:
A549 cell lysate at 20 µg

Secondary

Lanes 1 - 6:
Goat anti-Mouse IgG H&L 800CW at 1/20000 dilution

Lanes 1 - 6:
Goat anti-Rabbit IgG H&L 680RD at 1/20000 dilution

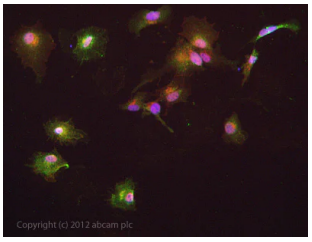
Observed band size: 52 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-IRAK4 antibody [2H9] (ab119942)

IRAK4 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-IRAK4 antibody

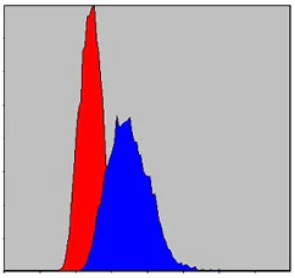
ab119942, at 1/200 dilution, staining IRAK4 in paraffin-embedded Human lung cancer tissue by Immunohistochemistry with DAB staining.



Immunocytochemistry/ Immunofluorescence - Anti-IRAK4 antibody [2H9] (ab119942)

IRAK4 Immunocytochemistry/ Immunofluorescence staining using mouse Anti-IRAK4 antibody

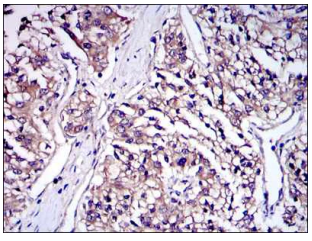
ab119942 stained HepG2 cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab119942 at 1/100 dilution overnight at +4°C. The secondary antibody (green) was DyLight® 488 goat anti- mouse (ab96879) IgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Flow Cytometry - Anti-IRAK4 antibody [2H9] (ab119942)

IRAK4 Flow Cytometry staining using mouse Anti-IRAK4 antibody

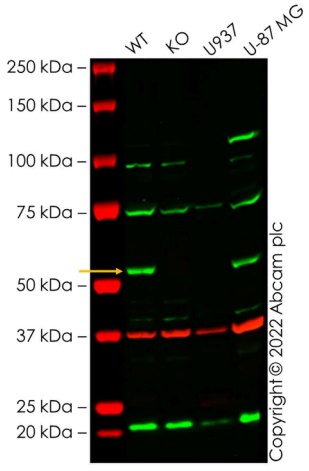
Flow cytometric analysis of Hela cells using ab119942 at 1/200 dilution (blue) and negative control (red).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-IRAK4 antibody [2H9] (ab119942)

IRAK4 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-IRAK4 antibody

ab119942, at 1/200 dilution, staining IRAK4 in paraffin-embedded Human kidney cancer tissue by Immunohistochemistry with DAB staining.



Western blot - Anti-IRAK4 antibody [2H9] (ab119942)

IRAK4 Western blot staining using mouse Anti-IRAK4 antibody

False colour image of Western blot: Anti-IRAK4 antibody [2H9] staining at 1/500 dilution, shown in green; Rabbit Anti-GAPDH antibody [EPR16891] (ab181602) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab119942 was shown to bind specifically to IRAK4. A band was observed at 55 kDa in wild-type THP-1 cell lysates with no signal observed at this size in IRAK4 knockout cell line ab281630 (knockout cell lysate ab282980). To generate this image, wild-type and IRAK4 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-Mouse IgG H&L (IRDye® 800CW) preabsorbed (ab216772) and Goat anti-Rabbit IgG H&L (IRDye® 680RD) preabsorbed (ab216777) at 1/20000 dilution.
All lanes:
Western blot - Anti-IRAK4 antibody [2H9] (ab119942) at 1/500 dilution

Lane 1:
Wild-type THP-1 cell lysate at 20 µg

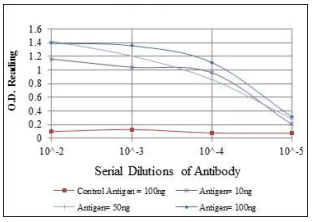
Lane 2:
IRAK4 knockout THP-1 cell lysate at 20 µg

Lane 3:
U937 cell lysate at 20 µg

Lane 4:
U-87 MG cell lysate at 20 µg

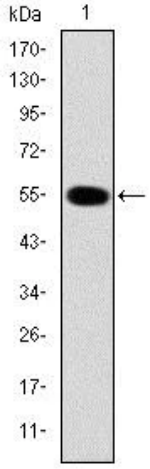
Performed under reducing conditions.

Observed band size: 55 kDa



ELISA - Anti-IRAK4 antibody [2H9] (ab119942)

ELISA using ab119942.

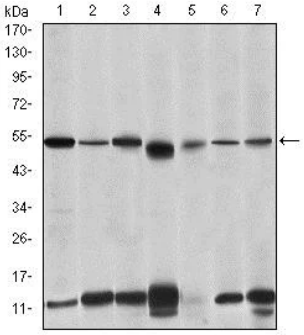


Western blot - Anti-IRAK4 antibody [2H9] (ab119942)

All lanes:
Western blot - Anti-IRAK4 antibody [2H9] (ab119942) at 1/500 dilution

All lanes:
Recombinant IRAK4 protein

Predicted band size: 52 kDa



Western blot - Anti-IRAK4 antibody [2H9] (ab119942)

All lanes:
Western blot - Anti-IRAK4 antibody [2H9] (ab119942) at 1/500 dilution

Lane 1:
THP-1 cell lysate

Lane 2:
HeLa cell lysate

Lane 3:
K562 cell lysate

Lane 4:
MCF7 cell lysate

Lane 5:
RAW264.7 cell lysate

Lane 6:
Jurkat cell lysate

Lane 7:
COS-7 (African green monkey kidney fibroblast-like cell line) cell lysate

Predicted band size: 52 kDa

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