abcam

ab224887

Anti-PKR antibody [EPR19374] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR19374
Purification technique	Affinity purification Protein A
Concentration	1.011 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

Species	Rat, Human
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Mouse, Human
Dilution info	-
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Mouse
Dilution info	-
Notes	-

Expected

Species	Rat, Human
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information EIF2AK2

Function

IFN-induced dsRNA-dependent serine/threonine-protein kinase that phosphorylates the alpha subunit of eukaryotic translation initiation factor 2 (EIF2S1/eIF-2-alpha) and plays a key role in the innate immune response to viral infection (PubMed:18835251, PubMed:19189853, PubMed:19507191, PubMed:21072047, PubMed:21123651, PubMed:22381929, PubMed:22948139, PubMed:23229543). Inhibits viral replication via the integrated stress response (ISR): EIF2S1/eIF-2-alpha phosphorylation in response to viral infection converts EIF2S1/eIF-2-alpha in a global protein synthesis inhibitor, resulting to a shutdown of cellular and viral protein synthesis, while concomitantly initiating the preferential translation of ISR-specific mRNAs, such as the transcriptional activator ATF4 (PubMed:19189853, PubMed:21123651, PubMed:22948139, PubMed:23229543). Exerts its antiviral activity on a wide range of DNA and RNA viruses including hepatitis C virus (HCV), hepatitis B virus (HBV), measles virus (MV) and herpes simplex virus 1 (HHV-1) (PubMed:11836380, PubMed:19189853, PubMed:19840259, PubMed:20171114, PubMed:21710204, PubMed:23115276, PubMed:23399035). Also involved in the regulation of signal transduction, apoptosis, cell proliferation and differentiation: phosphorylates other substrates including p53/TP53, PPP2R5A, DHX9, ILF3, IRS1 and the HHV-1 viral protein US11 (PubMed:11836380, PubMed:19229320, PubMed:22214662). In addition to serine/threonine-protein kinase activity, also has tyrosine-protein kinase activity and phosphorylates CDK1 at 'Tyr-4' upon DNA damage, facilitating its ubiquitination and proteasomal degradation (PubMed:20395957). Either as an adapter protein and/or via its kinase activity, can regulate various signaling pathways (p38 MAP kinase, NF-kappa-B and insulin signaling pathways) and transcription factors (JUN, STAT1, STAT3, IRF1, ATF3) involved in the expression of genes encoding pro-inflammatory cytokines and IFNs (PubMed:22948139, PubMed:23084476, PubMed:23372823). Activates the NF-kappa-B pathway via interaction with IKBKB and TRAF family of proteins and activates the p38 MAP kinase pathway via interaction with MAP2K6 (PubMed:10848580, PubMed:15121867, PubMed:15229216). Can act as both a positive and negative regulator of the insulin signaling pathway (ISP) (PubMed:20685959). Negatively regulates ISP by inducing the inhibitory phosphorylation of insulin receptor substrate 1 (IRS1) at 'Ser-312' and positively regulates ISP via phosphorylation of PPP2R5A which activates FOXO1, which in turn up-regulates the expression of insulin receptor substrate 2 (IRS2) (PubMed:20685959). Can regulate NLRP3 inflammasome assembly and the activation of NLRP3, NLRP1, AIM2 and NLRC4 inflammasomes (PubMed:22801494). Plays a role in the regulation of the cytoskeleton by binding to gelsolin (GSN), sequestering the protein in an inactive conformation away from actin (By similarity).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab224887 is the carrier-free version of ab184257.

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.

Conjugation ready

Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

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.

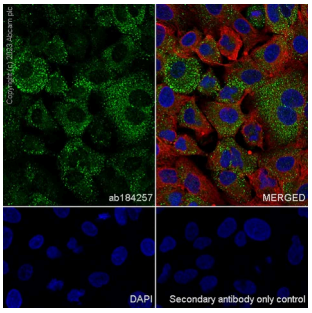
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

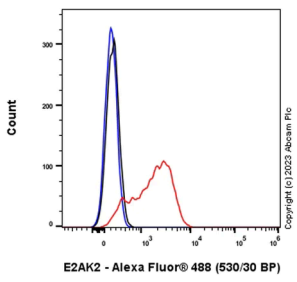
---

6 product images



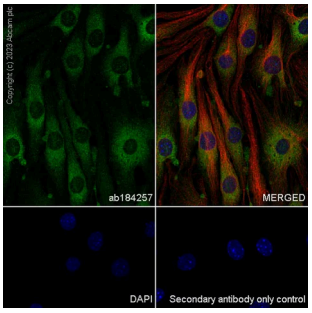
Immunocytochemistry/ Immunofluorescence - Anti-PKR antibody [EPR19374] - BSA and Azide free (ab224887)

This data was developed using ab184257, the same antibody clone in a different buffer formulation. Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A549 (human lung carcinoma epithelial cell) labeling PKR with ab184257 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preabsorbed (ab150081) secondary antibody at 1/1000 dilution (green). Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) (ab195889) at 1/200 was used as a counterstain The nuclear counterstain is DAPI (blue) Confocal image showing cytoplasmic and weak nuclear staining in A549 cell line. Image was taken with a confocal microscope(Leica-Microsystems, TCS SP8).



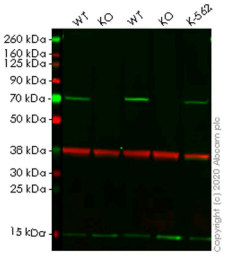
Flow Cytometry (Intracellular) - Anti-PKR antibody [EPR19374] - BSA and Azide free (ab224887)

This data was developed using ab184257, the same antibody clone in a different buffer formulation. Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methenol permeabilised bEnd.3 (mouse brain endothelial cell) cells labeling PKR with ab184257 at 1/500 dilution (red) compared with a Rabbit monoclonal IgG isotype control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-PKR antibody [EPR19374] - BSA and Azide free (ab224887)

This data was developed using ab184257, the same antibody clone in a different buffer formulation. Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized bEnd.3 (mouse brain endothelial cell) labeling PKR with ab184257 at 1/50 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preabsorbed (ab150081) secondary antibody at 1/1000 dilution (green). Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) (ab195889) at 1/200 was used as a counterstain The nuclear counterstain is DAPI (blue) Confocal image showing cytoplasmic and weak nuclear staining in bEnd.3 cell line. Image was taken with a confocal microscope(Leica-Microsystems, TCS SP8).



Western blot - Anti-PKR antibody [EPR19374] - BSA and Azide free (ab224887)

PKR Western blot staining using rabbit Anti-PKR antibody

This data was developed using ab184257, the same antibody clone in a different buffer formulation.
Lanes 1-5: Merged signal (red and green). Green - ab184257 observed at 70 kDa. Red - loading control ab8245 observed at 36 kDa.

ab184257 Anti-PKR antibody [EPR19374] was shown to specifically react with PKR in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab261824 (knockout cell lysate ab256899) was used. Wild-type and PKR knockout samples were subjected to SDS-PAGE. ab184257 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-PKR antibody [EPR19374] (ab184257) at 1/1000 dilution

Lane 1:
Wild-type HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

Lane 2:
EIF2AK2 knockout HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

Lane 2:
Western blot - Human EIF2AK2 (PKR) knockout HeLa cell line (ab261824)

Lane 3:
Wild-type A549 (Human lung carcinoma cell line) whole cell lysate at 20 µg

Lane 4:
EIF2AK2 knockout A549 (Human lung carcinoma cell line) whole cell lysate at 20 µg

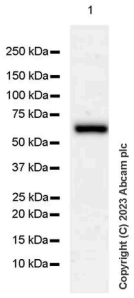
Lane 5:
K562 (Human chronic myelogenous leukemia lymphoblast cell line) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 62 kDa

Observed band size: 70 kDa



Western blot - Anti-PKR antibody [EPR19374] - BSA and Azide free (ab224887)

This data was developed using ab184257, the same antibody clone in a different buffer formulation. Blocking and diluting buffer: 5% NFDM/TBST"
All lanes:
Western blot - Anti-PKR antibody [EPR19374] (ab184257) at 1/1000 dilution

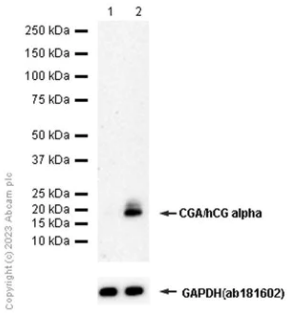
All lanes:
bEnd.3 (mouse brain endothelial cell) whole cell lysate at 20 µg

Secondary

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

Predicted band size: 62 kDa

Exposure time: 114s



Western blot - Anti-PKR antibody [EPR19374] - BSA and Azide free (ab224887)

This data was developed using ab184257, the same antibody clone in a different buffer formulation. Blocking and diluting buffer: 5% NFDM/TBST
Lanes 1 - 2:
Western blot - Anti-CGA/hCG alpha antibody [EP3373] (ab92738) at 1/2000 dilution

Lane 1:
Western blot - Anti-PKR antibody [EPR19374] (ab184257) at 1/1000 dilution

Lane 1:
JAR (Human placenta choriocarcinoma epithelial cell) whole cell lysate

Lane 1:
bEnd.3 (mouse brain endothelial cell) whole cell lysate at 20 µg

Lane 2:
JAR (Human placenta choriocarcinoma epithelial cell) whole cell lysate treated with Hydroxyurea (10mM 24h)

Secondary

Lanes 1 - 2:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

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

Performed under reducing conditions.

Observed band size: 18 kDa

Exposure time: 180s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.