abcam

Product datasheet

Anti-IRF1 antibody [EPR18301] ab186384

Recombinant RabMAb

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Overview

Product name	Anti-IRF1 antibody [EPR18301]
Description	Rabbit monoclonal [EPR18301] to IRF1
Host species	Rabbit
Tested applications	**Suitable for:** ICC/IF, WB, Flow Cyt, IP, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Recombinant fragment within Human IRF1 aa 100 to the C-terminus. The exact sequence is proprietary. Database link: P10914
Positive control	WB: Jurkat and IFN-gamma treated HeLa whole cell lysate; Mouse brain, heart and spleen lysates; rat brain and heart lysates; MOLT-4, C6, RAW 264.7, PC-12 and NIH/3T3 whole cell lysates. IHC-P: Human colon and Human gastric adenocarcinoma tissue. ICC/IF: MOLT-4, Jurkat, NIH\3T3, C6 cells. Flow Cytometry: Jurkat cells. IP: Jurkat 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. Reproducibility is key to advancing scientific discovery and accelerating scientists' next breakthrough. Abcam is leading the way with our range of recombinant antibodies, knockout-validated antibodies and knockout cell lines, all of which support improved reproducibility. We are also planning to innovate the way in which we present recommended applications and species on our product datasheets, so that only applications & species that have been tested in our own labs, our suppliers or by selected trusted collaborators are covered by our Abpromise™ guarantee. In preparation for this, we have started to update the applications & species that this product is Abpromise guaranteed for.

We are also updating the applications & species that this product has been "predicted to work with," however this information is not covered by our Abpromise guarantee.

Applications & species from publications and Abreviews that have not been tested in our own labs or in those of our suppliers are not covered by the Abpromise guarantee.

Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, as well as customer reviews and Q&As.

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
Clone number	EPR18301
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab186384** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

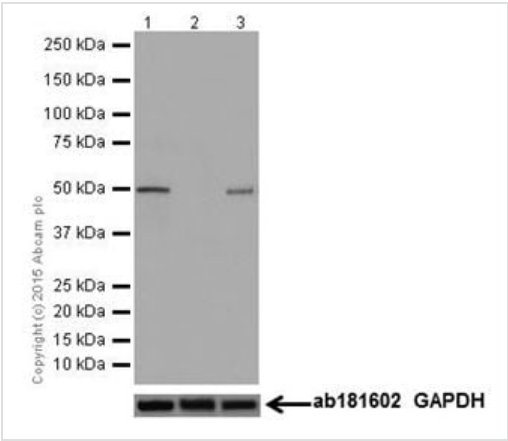
Application	Abreviews	Notes
ICC/IF		1/500.
WB	★★★☆☆	1/1000. Detects a band of approximately 48 kDa (predicted molecular weight: 37 kDa).
Flow Cyt		1/250.
IP		1/80.
IHC-P		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target

Function	Specifically binds to the upstream regulatory region of type I IFN and IFN-inducible MHC class I genes (the interferon consensus sequence (ICS)) and activates those genes. Acts as a tumor suppressor.

Involvement in disease	Defects in IRF1 are a cause of gastric cancer (GASC) [MIM:613659]; also called gastric cancer intestinal or stomach cancer. Gastric cancer is a malignant disease which starts in the stomach, can spread to the esophagus or the small intestine, and can extend through the stomach wall to nearby lymph nodes and organs. It also can metastasize to other parts of the body. The term gastric cancer or gastric carcinoma refers to adenocarcinoma of the stomach that accounts for most of all gastric malignant tumors. Two main histologic types are recognized, diffuse type and intestinal type carcinomas. Diffuse tumors are poorly differentiated infiltrating lesions, resulting in thickening of the stomach. In contrast, intestinal tumors are usually exophytic, often ulcerating, and associated with intestinal metaplasia of the stomach, most often observed in sporadic disease.
Sequence similarities	Belongs to the IRF family. Contains 1 IRF tryptophan pentad repeat DNA-binding domain.
Post-translational modifications	Sumoylation represses the transcriptional activity and displays enhanced resistance to protein degradation. Inactivates the tumor suppressor activity. Elevated levels in tumor cells. Major site is Lys-275. Sumoylation is enhanced by PIAS3 (By similarity). Desumoylated by SENP1 in tumor cells and appears to compete with ubiquitination on C-terminal sites. Ubiquitinated. Appears to compete with sumoylation on C-terminal sites.
Cellular localization	Nucleus.

Images



Western blot - Anti-IRF1 antibody [EPR18301] (ab186384)

All lanes : Anti-IRF1 antibody [EPR18301] (ab186384) at 1/10000 dilution

Lane 1 : Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysate
Lane 2 : Untreated HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate
Lane 3 : HeLa whole cell lysates treated with 10 ng/ml IFN-gamma for 24 hours

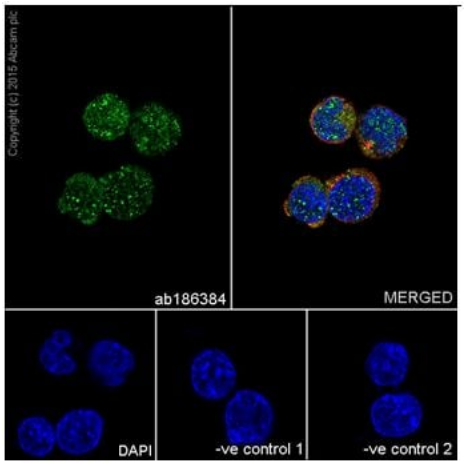
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 37 kDa
Observed band size: 48 kDa
why is the actual band size different from the predicted?

Exposure time: 15 seconds

Blocking/dilution buffer: 5% NFDM/TBST.



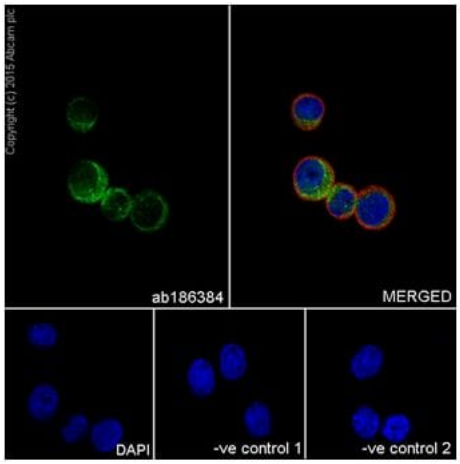
Immunocytochemistry/ Immunofluorescence - Anti-IRF1 antibody [EPR18301] (ab186384)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized MOLT-4 (Human lymphoblastic leukemia) cells labeling IRF1 with ab186384 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

Confocal image showing nuclear and cytoplasmic staining on MOLT-4 cells.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows:-
-ve control 1 - ab186384 at 1/500 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2. - ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



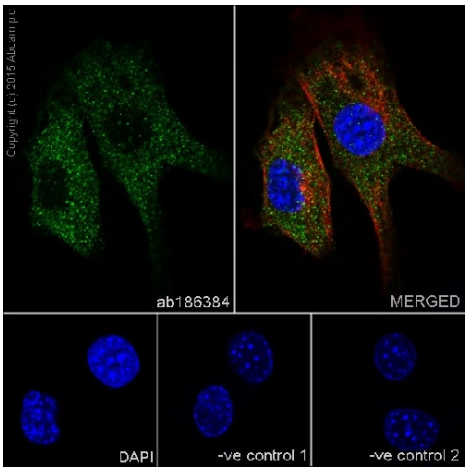
Immunocytochemistry/ Immunofluorescence - Anti-IRF1 antibody [EPR18301] (ab186384)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Jurkat (Human T cell leukemia from peripheral blood) cells labeling IRF1 with ab186384 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

Confocal image showing cytoplasmic and weakly nuclear staining on Jurkat cells.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows:-
-ve control 1 - ab186384 at 1/500 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2. - ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



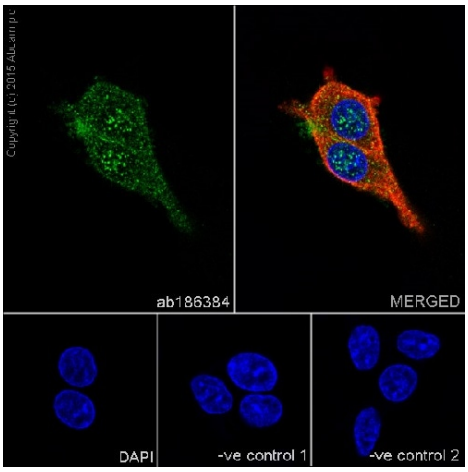
Immunocytochemistry/ Immunofluorescence - Anti-IRF1 antibody [EPR18301] (ab186384)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embyro fibroblast cells) cells labeling IRF1 with ab186384 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

Confocal image showing cytoplasmic and weakly nuclear staining on NIH/3T3 cells.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows:-
-ve control 1 - ab186384 at 1/500 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2. - ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



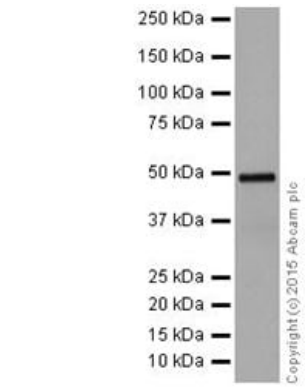
Immunocytochemistry/ Immunofluorescence - Anti-IRF1 antibody [EPR18301] (ab186384)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized C6 (Rat glial tumor cells) cells labeling IRF1 with ab186384 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green).

Confocal image showing cytoplasmic and nuclear staining on C6 cells.

The nuclear counterstain is DAPI (blue).

Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).
The negative controls are as follows:-
-ve control 1 - ab186384 at 1/500 dilution followed by ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
-ve control 2. - ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150077 (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.



Western blot - Anti-IRF1 antibody [EPR18301] (ab186384)

Anti-IRF1 antibody [EPR18301] (ab186384) at 1/1000 dilution + MOLT-4 (Human lymphoblastic leukemia) whole cell lysate 20ug

Secondary

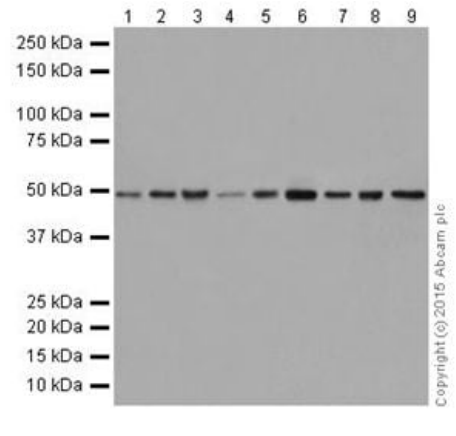
Goat Anti-Rabbit IgG, (H+L),Peroxidase conjugated at 1/1000 dilution

Predicted band size: 37 kDa

Observed band size: 48 kDa why is the actual band size different from the predicted?

Exposure time: 30 seconds

Blocking/dilution buffer: 5% NFDM/TBST.



Western blot - Anti-IRF1 antibody [EPR18301] (ab186384)

All lanes : Anti-IRF1 antibody [EPR18301] (ab186384) at 1/1000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse heart lysate
Lane 3 : Mouse spleen lysate
Lane 4 : Rat brain lysate
Lane 5 : Rat heart lysate
Lane 6 : C6 (rat glioma tumor) whole cell lysate
Lane 7 : RAW 264.7 (mouse macrophage cells transformed with Abelson murine leukemia virus) whole cell lysate
Lane 8 : PC-12 (rat adrenal gland pheochromocytoma) whole cell lysate
Lane 9 : NIH/3T3 (mouse embryo fibroblast) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

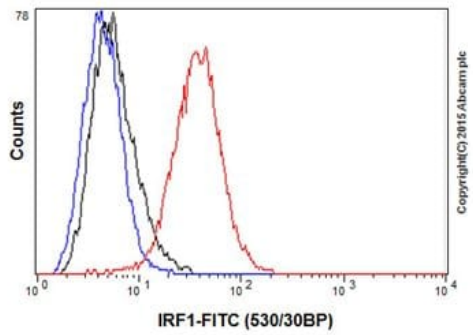
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 37 kDa

Observed band size: 48 kDa why is the actual band size different from the predicted?

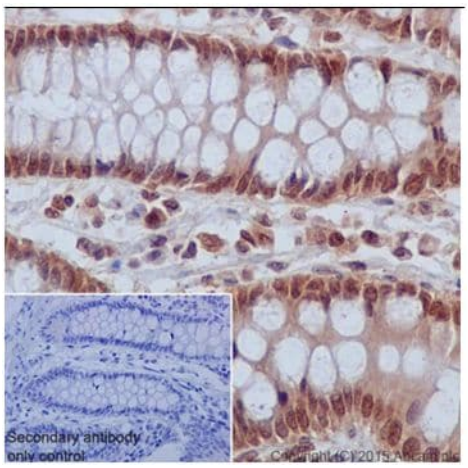
Exposure time: 30 seconds

Blocking/dilution buffer: 5% NFDM/TBST.



Flow Cytometry - Anti-IRF1 antibody [EPR18301] (ab186384)

Flow cytometric analysis of 2% paraformaldehyde-fixed Jurkat (Human T cell leukemia cells fromperipheral blood) cells labeling IRF1 with ab186384 at 1/250 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 (FITC) at 1/150 dilution was used as the secondary antibody.



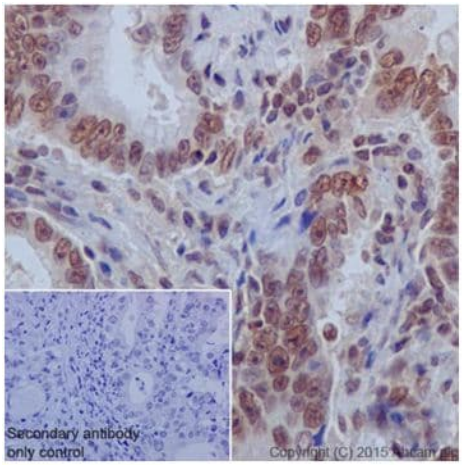
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-IRF1 antibody [EPR18301] (ab186384)

Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling IRF1 with ab186384 at 1/500 dilution followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Nuclear and cytoplasmic staining on epithelial cells of normal Human colon is observed.

Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary ab, secondary ab is Goat Anti-Rabbit IgG H&L (HRP) (ab97051).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



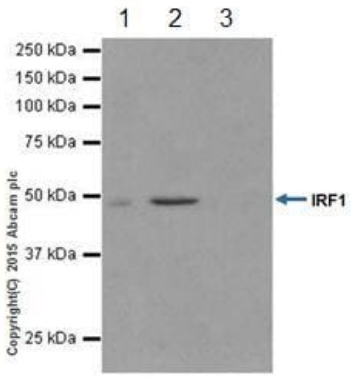
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-IRF1 antibody [EPR18301] (ab186384)

Immunohistochemical analysis of paraffin-embedded Human gastric adenocarcinoma tissue labeling IRF1 with ab186384 at 1/500 dilution followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Nuclear and weak cytoplasmic staining on tumor cells of gastric adenocarcinoma is observed.

Counter stained with Hematoxylin.
Negative control: Used PBS instead of primary ab, secondary ab is Goat Anti-Rabbit IgG H&L (HRP) (ab97051).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-IRF1 antibody [EPR18301] (ab186384)

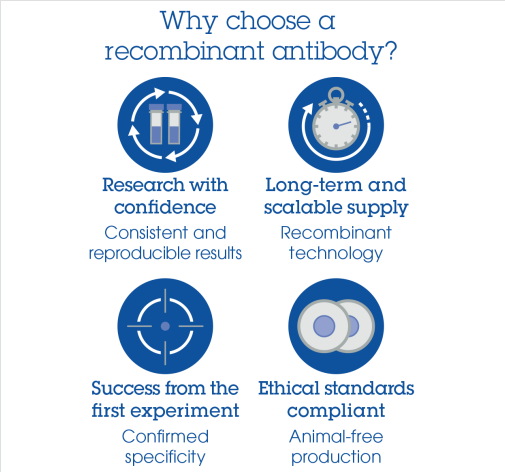
IRF1 was immunoprecipitated from 1mg of Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysate with ab186384 at 1/80 dilution.

Lane 1: Input Jurkat whole cell extract (10µg).

Lane 2: Jurkat whole cell lysate following immunoprecipitation with ab186384.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab186384 in Jurkat whole cell lysate.

Western blot was performed from the immunoprecipitate using ab186384 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1/1500 dilution.
Blocking and dilution buffer and concentration: 5% NFDM/TBST. 30 second exposure.



Anti-IRF1 antibody [EPR18301] (ab186384)

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