abcam

Product datasheet

Anti-ATF5 antibody [EPR18286] ab184923

Recombinant RabMAb

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Overview

Product name	Anti-ATF5 antibody [EPR18286]
Description	Rabbit monoclonal [EPR18286] to ATF5
Host species	Rabbit
Tested applications	**Suitable for:** WB, IHC-P, ICC/IF, IP, Flow Cyt (Intra)
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Jurkat, HEK-293, SH-SY5Y, Neuro-2a, C6, PC-12, RAW 264.7 and NIH/3T3 whole cell lysates; Human fetal brain, fetal heart and fetal kidney lysates; Rat and mouse brain, heart and kidney lysates. IHC-P: Human breast, Human hepatocellular carcinoma, mouse cardiac muscle and rat stomach tissues. ICC/IF: NIH/3T3 and Jurkat cells. IP: NIH/3T3 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	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR18286

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab184923 in the following tested applications.

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

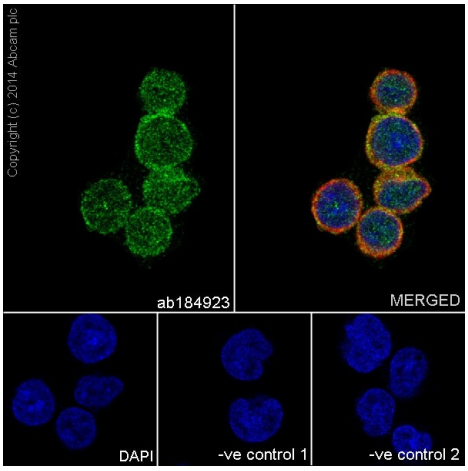
Application	Abreviews	Notes
WB	★★★★☆ (1)	1/2000. Detects a band of approximately 31 kDa (predicted molecular weight: 31 kDa).
IHC-P		1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/500.
IP		1/50.
Flow Cyt (Intra)		Use at an assay dependent concentration.

Target

Relevance ATF5 or Activating transcription factor 5, binds to cAMP inducible promoters and is involved in gene transcription. This protein binds the cAMP response element (CRE) (consensus: 5'-GTGACGT[AC][AG]-3'), a sequence present in many viral and cellular promoters. ATF5 plays a role in inhibition of nerve growth factor induced neuronal outgrowth and regulation of neurogenesis.

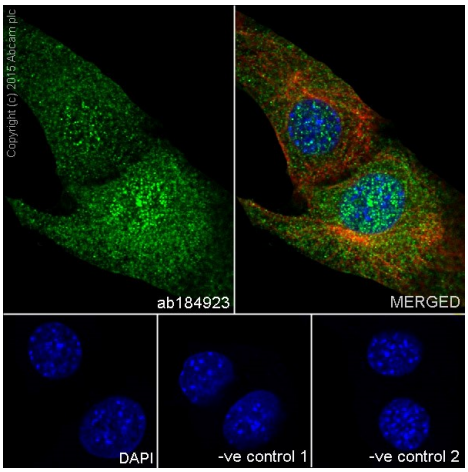
Cellular localization Cytoplasmic and Nuclear

Images



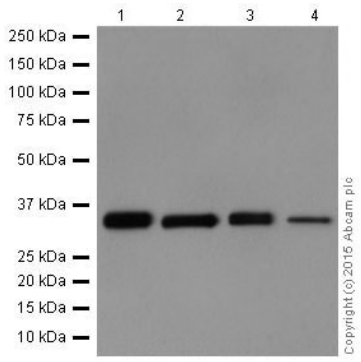
Immunocytochemistry/ Immunofluorescence - Anti-ATF5 antibody [EPR18286] (ab184923)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Jurkat (Human T cell leukemia cells from peripheral blood) cells labeling ATF5 with ab184923 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear and cytoplasmic staining on Jurkat cell line. The nuclear counter stain 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 1/1000 dilution (red).
The negative controls are as follows;
-ve control 1: ab184923 at 1/500 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 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 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-ATF5 antibody [EPR18286] (ab184923)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embyro fibroblast cells) cells labeling ATF5 with ab184923 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear and cytoplasmic staining on NIH/3T3 cell line. The nuclear counter stain 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 1/1000 dilution (red).
The negative controls are as follows;
-ve control 1: ab184923 at 1/500 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/1000 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 1/1000 dilution.



Western blot - Anti-ATF5 antibody [EPR18286] (ab184923)

All lanes : Anti-ATF5 antibody [EPR18286] (ab184923) at 1/2000 dilution

Lane 1 : Jurkat (Human T cell leukemia cells from peripheral blood) whole cell lysate
Lane 2 : HEK-293 (Human epithelial cells from embryonic kidney) whole cell lysate
Lane 3 : SH-SY5Y (Human neuroblastoma from bone marrow cells) whole cell lysate
Lane 4 : Neuro-2a (Mouse neuroblastoma cells) whole cell lysate

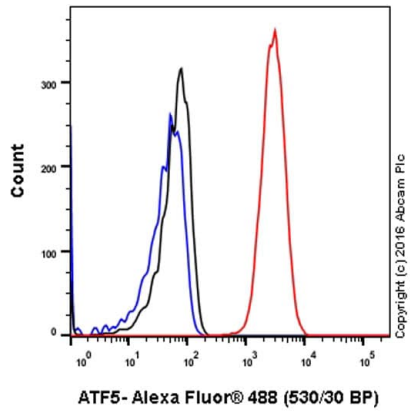
Lysates/proteins at 20 µg per lane.

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

Predicted band size: 31 kDa
Observed band size: 31 kDa

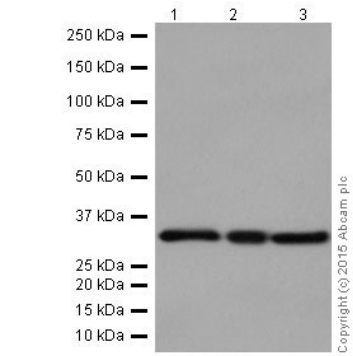
Exposure time: 10 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Flow Cytometry (Intracellular) - Anti-ATF5 antibody [EPR18286] (ab184923)

Intracellular Flow Cytometry analysis of Jurkat (human acute T cell leukemia) cells labeling ATF5 with purified ab184923 at 1/120 dilution (10ug/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) was used as the unlabeled control.



Western blot - Anti-ATF5 antibody [EPR18286] (ab184923)

All lanes : Anti-ATF5 antibody [EPR18286] (ab184923) at 1/2000 dilution

Lane 1 : Human fetal brain lysate
Lane 2 : Human fetal heart lysate
Lane 3 : Human fetal kidney lysate

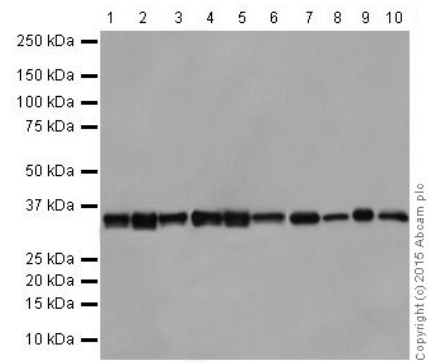
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 31 kDa
Observed band size: 31 kDa

Exposure time: 10 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.



Western blot - Anti-ATF5 antibody [EPR18286] (ab184923)

All lanes : Anti-ATF5 antibody [EPR18286] (ab184923) at 1/2000 dilution

Lane 1 : Mouse brain lysate
Lane 2 : Mouse heart lysate
Lane 3 : Mouse kidney lysate
Lane 4 : Rat brain lysate
Lane 5 : Rat heart lysate
Lane 6 : Rat kidney lysate
Lane 7 : C6 (Rat glial tumor cells) whole cell lysate
Lane 8 : RAW 264.7 (Mouse macrophage cells transformed with Abelson murine leukemia virus) whole cell lysate
Lane 9 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysate
Lane 10 : NIH/3T3 (Mouse embyro fibroblast cells) whole cell lysate

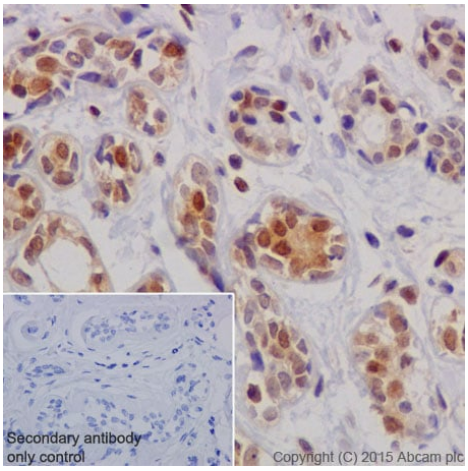
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab136636**) at 1/1000 dilution

Predicted band size: 31 kDa
Observed band size: 31 kDa

Exposure time: 10 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

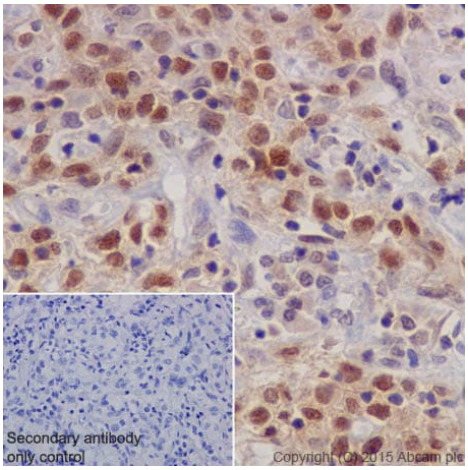


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATF5 antibody [EPR18286] (ab184923)

Immunohistochemical analysis of paraffin-embedded Human breast tissue labeling ATF5 with ab184923 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nucleus and weak cytoplasm staining on Human breast is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

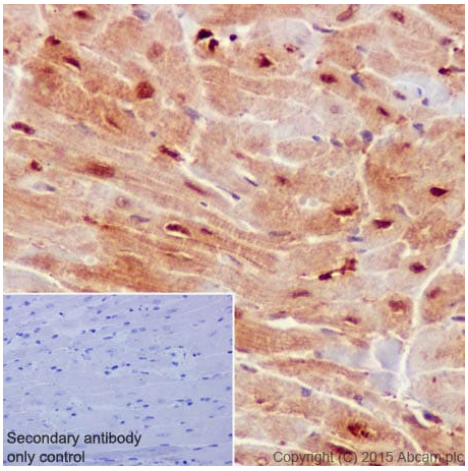


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATF5 antibody [EPR18286] (ab184923)

Immunohistochemical analysis of paraffin-embedded Human hepatocellular carcinoma tissue labeling ATF5 with ab184923 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nucleus and weak cytoplasm staining on tumor cells of hepatocellular carcinoma is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

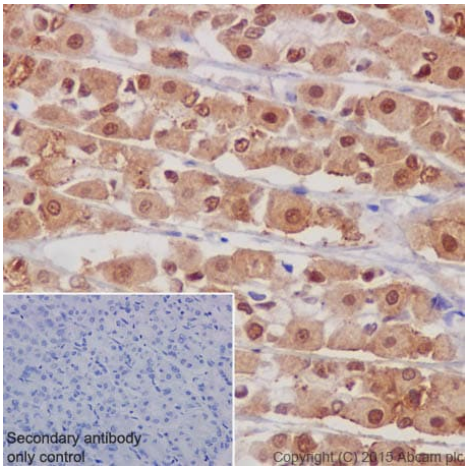


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATF5 antibody [EPR18286] (ab184923)

Immunohistochemical analysis of paraffin-embedded Mouse cardiac muscle tissue labeling ATF5 with ab184923 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nucleus and cytoplasm staining on mouse cardiac muscle is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

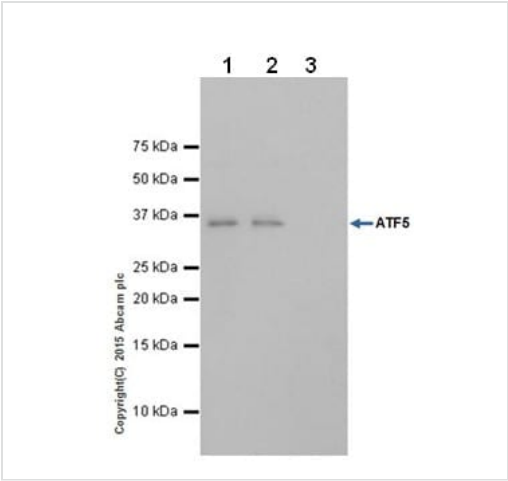


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATF5 antibody [EPR18286] (ab184923)

Immunohistochemical analysis of paraffin-embedded Rat stomach tissue labeling ATF5 with ab184923 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) secondary antibody at 1/500 dilution. Nucleus and cytoplasm staining on rat stomach is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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

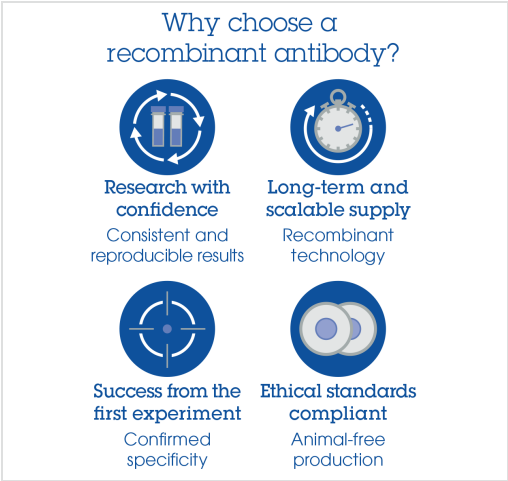


Immunoprecipitation - Anti-ATF5 antibody [EPR18286] (ab184923)

ATF5 was immunoprecipitated from 1mg of NIH/3T3 (Mouse embyro fibroblast cells) whole cell lysate with ab184923 at 1/50 dilution. Western blot was performed from the immunoprecipitate using ab184923 at 1/10000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1/1500 dilution.

Lane 1: NIH/3T3 whole cell lysate 10 µg (Input). Lane 2: ab184923 IP in NIH/3T3 whole cell lysate. Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of ab184923 in NIH/3T3 whole cell lysate. Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 1 second



Anti-ATF5 antibody [EPR18286] (ab184923)

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