# abcam

### Product datasheet

# Anti-ATF2 antibody [EPR22938-114] ab239361

Recombinant RabMAb

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Overview

**Product name** Anti-ATF2 antibody [EPR22938-114]

**Description** Rabbit monoclonal [EPR22938-114] to ATF2

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), WB, IP, IHC-P

Unsuitable for: ChIP or ICC/IF

Species reactivity Reacts with: Mouse. Rat. Human

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

Positive control WB: NIH/3T3, RAW 264.7, HEK-293T, K562, PC-12 and Jurkat whole cell lysate. Mouse brain

> tissue lysate. Rat spleen tissue lysate. IHC-P: Human kidney carcinoma tissue. Human, mouse and rat kidney tissue. Flow Cyt (intra): RAW 264.7 and NIH/3T3 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: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal

Clone number EPR22938-114

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab239361 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
Flow Cyt (Intra)		1/40.
WB		1/1000.
IP		1/30.
IHC-P		1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

**Application notes** Is unsuitable for ChIP or ICC/IF.

**Target** 

Function Transcriptional activator, probably constitutive, which binds to the cAMP-responsive element

(CRE) (consensus: 5'-GTGACGT[AC][AG]-3'), a sequence present in many viral and cellular promoters. Interaction with JUN redirects JUN to bind to CRES preferentially over the 12-O-tetradecanoylphorbol-13-acetate response elements (TRES) as part of an ATF2/JUN complex.

**Tissue specificity** Abundant expression seen in the brain.

**Sequence similarities** Belongs to the bZIP family. ATF subfamily.

Contains 1 bZIP domain.

Contains 1 C2H2-type zinc finger.

Post-translational

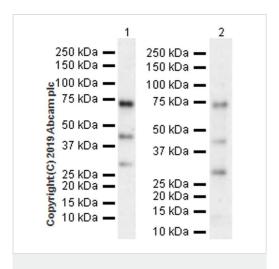
modifications

Phosphorylation of Thr-69 and Thr-71 by MAPK14 causes increased transcriptional activity. Also

phosphorylated and activated by JNK.

Cellular localization Nucleus.

#### **Images**



Western blot - Anti-ATF2 antibody [EPR22938-114] (ab239361)

**All lanes :** Anti-ATF2 antibody [EPR22938-114] (ab239361) at 1/1000 dilution

**Lane 1**: PC-12 (rat adrenal gland pheochromocytoma), whole cell lysate

Lane 2: Rat spleen tissue lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

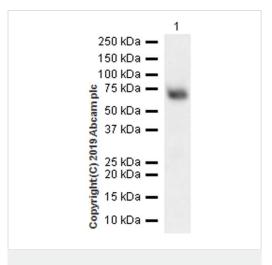
**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Observed band size: 70 kDa

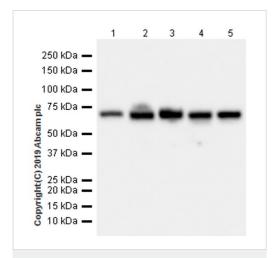
The molecular weight and degraded fragments observed are consistent with what has been described in the literature (PMID: 9488727, 10207054, 26901653). Suggest use freshly made lysate to minimize protein degradation.

Exposure time: 3 minutes.

Blocking and Diluting Buffer and concentration: 5% NFDM/TBST.



Western blot - Anti-ATF2 antibody [EPR22938-114] (ab239361)



Western blot - Anti-ATF2 antibody [EPR22938-114] (ab239361)

Anti-ATF2 antibody [EPR22938-114] (ab239361) at 1/1000 dilution

+ Mouse brain tissue lysate at 20 µg

#### Secondary

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

Observed band size: 70 kDa

The molecular weight observed is consistent with what has been described in the literature (PMID:26901653).

Exposure time: 3 minutes.

Blocking and Diluting Buffer and concentration: 5% NFDM/TBST.

**All lanes :** Anti-ATF2 antibody [EPR22938-114] (ab239361) at 1/1000 dilution

Lane 1: NIH/3T3 (mouse embryonic fibroblast), whole cell lysate

Lane 2: RAW264.7 (mouse Abelson murine leukemia virus-

induced tumor macrophage), whole cell lysate

**Lane 3**: HEK-293T (human embryonic kidney epithelial cell), whole cell lysate

**Lane 4**: K562 (human chronic myelogenous leukemia lymphoblast), whole cell lysate

Lane 5 : Jurkat (human T cell leukemia T lymphocyte), whole cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

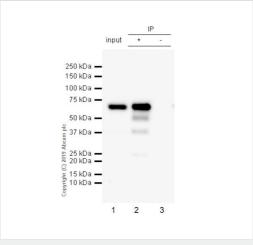
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

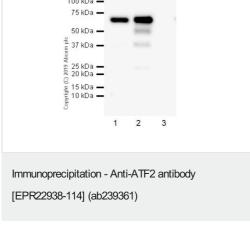
Observed band size: 70 kDa

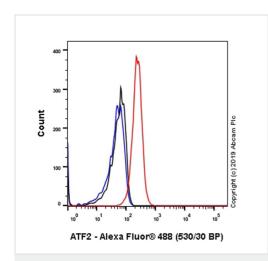
The molecular weight observed is consistent with what has been described in the literature (PMID:26901653).

Exposure time: 10 seconds.

Blocking and Diluting Buffer and concentration: 5% NFDM/TBST.







Flow Cytometry (Intracellular) - Anti-ATF2 antibody [EPR22938-114] (ab239361)

ATF2 was immunoprecipitated from 0.35 mg NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate with ab239361 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab239361. VeriBlot for IP Detection Reagent (HRP) (ab131366) was used at 1/1000 dilution.

Lane 1: NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate 10µg.

Lane 2: ab239361 IP in NIH/3T3 whole cell lysate.

Lane 3: Rabbit monoclonal lgG (ab172730) instead of ab239361 in NIH/3T3 whole cell lysate.

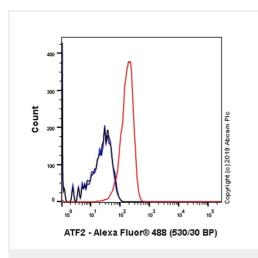
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 8 seconds

Lysate were made freshly and used in IP test immediately to minimize protein degradation. Incubation time was 2h.

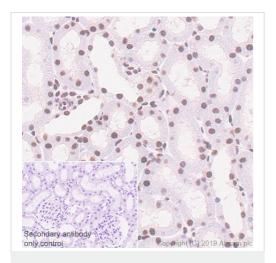
The molecular weight and degraded fragments observed are consistent with what has been described in the literature (PMID: 9488727, PMID: 10207054, PMID:26901653).

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (Mouse embryonic fibroblast) cells labeling ATF2 with ab239361 at 1/40 (Red) compared with a Rabbit monoclonal lgG (ab172730) (Black) isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit lgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



Flow Cytometry (Intracellular) - Anti-ATF2 antibody [EPR22938-114] (ab239361)

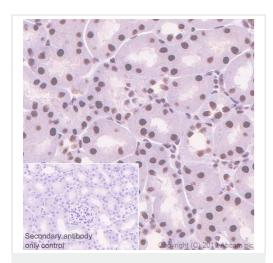
Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) cells labeling ATF2 with ab239361 at 1/40 (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATF2 antibody
[EPR22938-114] (ab239361)

Immunohistochemical analysis of paraffin-embedded rat kidney tissue labeling ATF2 with ab239361 at 1/250 dilution (1.76 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Nuclear staining on rat kidney is observed. The section was incubated with ab239361 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20mins.

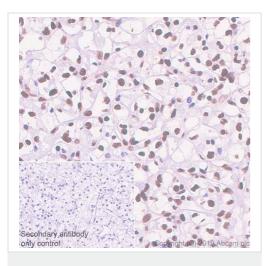
Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATF2 antibody
[EPR22938-114] (ab239361)

Immunohistochemical analysis of paraffin-embedded mouse kidney tissue labeling ATF2 with ab239361 at 1/250 dilution (1.76 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Nuclear staining on mouse kidney is observed. The section was incubated with ab239361 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20mins.

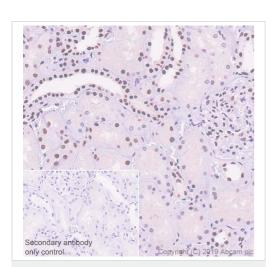
Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATF2 antibody
[EPR22938-114] (ab239361)

Immunohistochemical analysis of paraffin-embedded human kidney carcinoma tissue labeling ATF2 with ab239361 at 1/250 dilution (1.76 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<a href="mailto:ab209101">ab209101</a>). Nuclear staining on human kidney carcinoma (PMID: 27377902) is observed. The section was incubated with ab239361 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20mins.

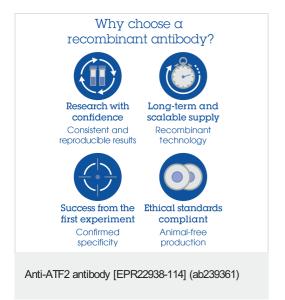
Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-ATF2 antibody
[EPR22938-114] (ab239361)

Immunohistochemical analysis of paraffin-embedded human kidney tissue labeling ATF2 with ab239361 at 1/250 dilution (1.76 ug/ml) followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Nuclear staining on human kidney (PMID: 27377902) is observed. The section was incubated with ab239361 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with hematoxylin. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20mins.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>).



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