abcam

Product datasheet

Anti-ATF2 antibody [E243] - ChIP Grade ab32160

Recombinant RabMAb

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Overview

Product name	Anti-ATF2 antibody [E243] - ChIP Grade
Description	Rabbit monoclonal [E243] to ATF2 - ChIP Grade
Host species	Rabbit
Specificity	This antibody recognises ATF2, but does not cross react with other ATF family members.
Tested applications	**Suitable for:** Flow Cyt (Intra), ChIP, WB, IHC-P, IP, ICC/IF
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide within Human ATF2 aa 450-550 (C terminal). The exact sequence is proprietary.
Positive control	WB: HeLa cell lysate. IHC-P: Breast carcinoma tissue. Flow Cyt (intra): HeLa cells. IP: HeLa cell lysate. ChIP: Jurkat cells. ICC/IF: A549 cells.
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**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 49% PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA
Clonality	Monoclonal

Clone number	E243
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab32160 in the following tested applications.

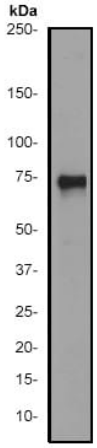
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/2000.
ChIP		Use 5 µg for 25 µg of chromatin.
WB	★★★☆☆ (1)	1/10000. Detects a band of approximately 70 kDa (predicted molecular weight: 54 kDa).
IHC-P	★★★★★ (1)	1/250 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
IP		1/60.
ICC/IF		1/250 - 1/500.

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.

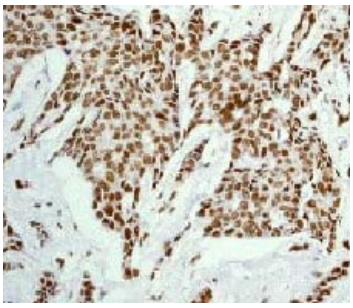
Images



Western blot - Anti-ATF2 antibody [E243] - ChIP Grade (ab32160)

Anti-ATF2 antibody [E243] - ChIP Grade (ab32160) at 1/10000 dilution + HeLa cell lysate

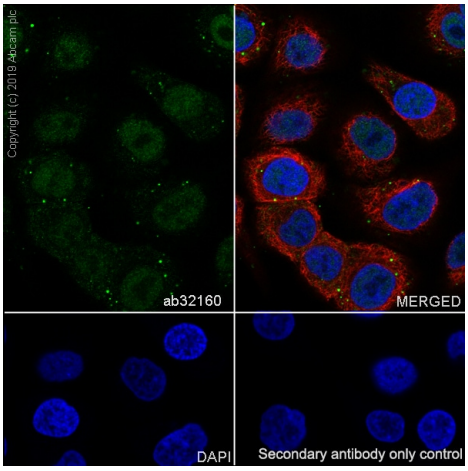
Predicted band size: 54 kDa
Observed band size: 70 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATF2 antibody [E243] - ChIP Grade (ab32160)

Ab32160, at a dilution of 1/250, staining ATF2 in paraffin embedded breast carcinoma tissue by Immunohistochemistry.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

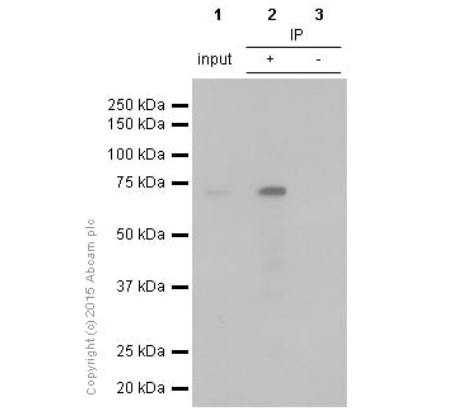


Immunocytochemistry/ Immunofluorescence - Anti-ATF2 antibody [E243] - ChIP Grade (ab32160)

Immunocytochemistry analysis of A549 (human lung carcinoma epithelial cell) labeling ATF2 with purified ab32160 at 1/100 dilution (10 µg/ml). Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) at 1/1000 (2 µg/ml) was used as the secondary antibody. **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.10 µg/ml) was used as counterstain. Nuclei were stained blue with DAPI.
Negative control: PBS instead of the primary antibody.

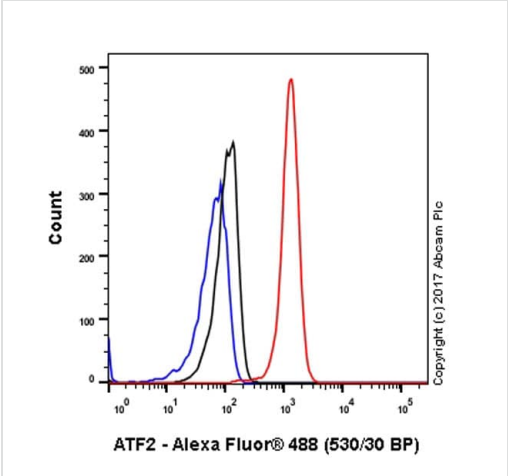
ChIP - Anti-ATF2 antibody [E243] - ChIP Grade (ab32160)

Chromatin was prepared from Jurkat (TPA and Ionomycin treated or not) cells according to the Abcam X-ChIP protocol. Cells were fixed with 1% formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 5µg of ab32160 (red), and 20µl of protein A/G sepharose beads slurry (10µl of sepharose A beads + 10µl of sepharose G beads). 5µg of rabbit normal IgG was added to the beads control (grey). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach).



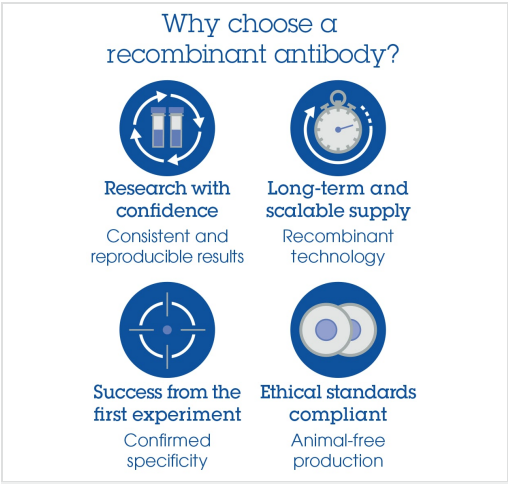
Immunoprecipitation - Anti-ATF2 antibody [E243] - ChIP Grade (ab32160)

ab32160 (purified) at 1/60 dilution (20 µg/mL) immunoprecipitating ATF2 in HeLa whole cell lysate.
Lane 3 (-): HeLa(Human cervix adenocarcinoma epithelial cell) whole cell lysate 10µgab32160 & HeLa whole cell lysateRabbit monoclonal IgG (**ab172730**) instead of ab32160 in HeLa whole cell lysate
For western blotting, ab32160 at 1/1000 dilution (2.284 µg/mL) and bbit TureBlot: Anti-Rabbit IgG HRP was used as the secondary antibody at 1/1500 dilution.
Blocking and diluting buffer: 5% NFDM /TBST .



Flow Cytometry (Intracellular) - Anti-ATF2 antibody [E243] - ChIP Grade (ab32160)

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling ATF2 (red) with ab32160 at a 1/2000 dilution. Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A goat anti-rabbit IgG (Alexa Fluorr® 488) (**ab150077**) was used as the secondary antibody at a 1/2000 dilution. Black - Rabbit monoclonal IgG (**ab172730**). Blue (unlabeled control) - Cells without incubation with the primary and secondary antibodies.



Anti-ATF2 antibody [E243] - ChIP Grade (ab32160)

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