abcam

Product datasheet

Anti-ATF2 (phospho T71) antibody [E268] ab32019

יעלאעבע RabMAb

9 References 画像数7

製品の概要

製品名 Anti-ATF2 (phospho T71) antibody [E268]

製品の詳細 Rabbit monoclonal [E268] to ATF2 (phospho T71)

由来種 Rabbit

アプリケーション 適用あり: WB, IP, ICC/IF, Dot blot

適用なし: Flow Cyt or IHC

種交差性 交差種: Mouse, Human

免疫原 Synthetic peptide corresponding to Human ATF2 aa 50-150.

Database link: P15336

ポジティブ・コントロール IP: HeLa treated with 250ng/ml anisomycin for 30min whole cell lysate. ICC/IF: HeLa cells treated

with 250ng/ml anisomycin for 30min.

特記事項 SAPK and p38 MAPK activate, in response to cellular stress, ATF2 by phosphorylating the

protein at Thr69 and Thr71. Mutations of these sites result in the loss of stress induced

transcription by ATF2.

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**.

Rat: We have preliminary internal testing data to indicate this antibody may not react with this

species. Please contact us for more information.

製品の特性

製品の状態 Liquid

保存方法 Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

バッファー pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 49% PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA

精製度 Protein A purified

一次抗体 備考 SAPK and p38 MAPK activate, in response to cellular stress, ATF2 by phosphorylating the

protein at Thr69 and Thr71. Mutations of these sites result in the loss of stress induced

transcription by ATF2.

ポリ/モノ モノクローナル

クローン名 E268 アイソタイプ IgG

アプリケーション

The Abpromise guarantee Abpromise保証は、次のテスト済みアプリケーションにおけるab32019の使用に適用されますアプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		1/5000. Detects a band of approximately 70 kDa (predicted molecular weight: 54 kDa).
IP		1/30.
ICC/IF		1/500.
Dot blot		Use at an assay dependent concentration.

追加情報 Is unsuitable for Flow Cyt or IHC.

ターゲット情報

機能

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-Otetradecanoylphorbol-13-acetate response elements (TRES) as part of an ATF2/JUN complex.

組織特異性 Abundant expression seen in the brain.

配列類似性 Belongs to the bZIP family. ATF subfamily.

Contains 1 bZIP domain.

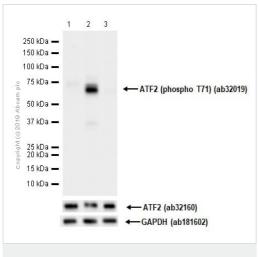
Contains 1 C2H2-type zinc finger.

翻訳後修飾 Phosphorylation of Thr-69 and Thr-71 by MAPK14 causes increased transcriptional activity. Also

phosphorylated and activated by JNK.

細胞内局在 Nucleus.

画像



Western blot - Anti-ATF2 (phospho T71) antibody [E268] (ab32019)

All lanes : Anti-ATF2 (phospho T71) antibody [E268] (ab32019) at 0.526 µg/ml

Lane 1: HeLa (human cervix adenocarcinoma epithelial cell) grown in serum free media for overnight, whole cell lysate

Lane 2: HeLa grown in serum free media for overnight, then treated with 250ng/ml Anisomycin for 30min, whole cell lysate

Lane 3: HeLa grown in serum free media for overnight, then treated with 250ng/ml Anisomycin for 30min whole cell lysate. Then the membrane was incubated with alkaline phosphatase.

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) ($\underline{ab97051}$) at 0.05 $\mu g/ml$

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

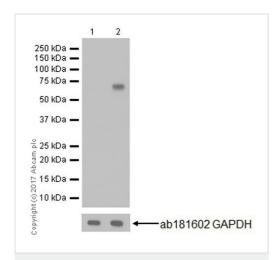
ab32019 DAPI MERGED

AB032019 DAPI MERGED

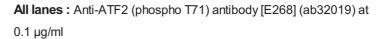
Immunocytochemistry/ Immunofluorescence - Anti-ATF2 (phospho T71) antibody [E268] (ab32019)

Blocking/Diluting Buffer and concentration: $5\%\ NFDM\ / TBST$

Immunocytochemistry of HeLa (Human epithelial cell line from cervix adenocarcinoma), prepared in FBS free medium overnight labeling ATF2 at 0.9 μ g/ml. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% tritonX-100. AlexaFluor[®]488 Goat anti-Rabbit (ab150077) was used as the secondary antibody at 1/500 . Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor[®] 594) (ab195889) was used as the counter stain at 2.5 μ g/ml. DAPI was used for nuclear counter stain. Confocal image showing the expression was increased on HeLa cells, prepared in FBS free medium overnight, then treated with 250ng/ml anisomycin for 30min.



Western blot - Anti-ATF2 (phospho T71) antibody [E268] (ab32019)



Lane 1 : Untreated NIH/3T3 (mouse embryo), prepared in FBS free medium overnight, whole cell lysate

Lane 2: NIH/3T3, prepared in FBS free medium overnight, then treated with 10 µg/ml anisomycin for 30 minutes whole cell lysate

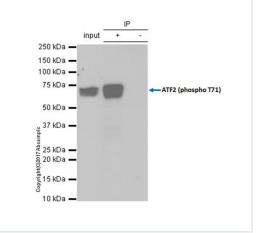
Lysates/proteins at 10 µg per lane.

Secondary

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

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

Exposure time: 5 seconds



Immunoprecipitation - Anti-ATF2 (phospho T71) antibody [E268] (ab32019)

Blocking and diluting buffer used was 2% BSA/TBST.

Lane 1 (input): HeLa (human cervix adenocarcinoma) treated with 250ng/ml anisomycin for 30min whole cell lysate 10µg.

Lane 2 (+): HeLa treated with 250ng/ml anisomycin for 30min whole cell lysate.

Lane 3 (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab32019 in HeLa treated with 250ng/ml anisomycin for 30min whole cell lysate.

Ab32019 Immunoprecipitating ATF2 in Human Hela whole cell lysate. For western blotting ab32019 (1:1000) was used to confirm successful immunoprecipitation. Blocking and diluting buffer used was 5% NFDM/TBST.

All lanes : Anti-ATF2 (phospho T71) antibody [E268] (ab32019) at 0.47 $\mu g/ml$

Lane 1: HeLa (human cervix adenocarcinoma) treated with 250ng/ml anisomycin for 30min whole cell lysate at 10 µg

Lane 2: HeLa treated with 250ng/ml anisomycin for 30min whole cell lysate

Lane 3 : Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab32019 in HeLa treated with 250ng/ml anisomycin for 30min whole cell lysate

Secondary

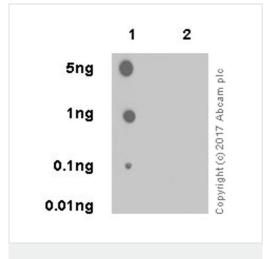
All lanes : VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/10000 dilution

Exposure time: 3 minutes

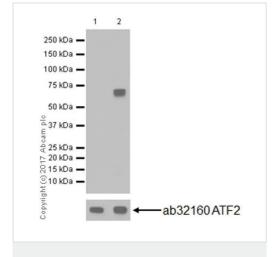
Dot blot analysis of ATF2 (phospho T71) peptide (Lane 1) and ATF2 non-phospho peptide (Lane 2) using ab32019 at 1/1000 dilution followed by Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/100000 dilution.

Exposure time: 3 minutes

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



Dot Blot - Anti-ATF2 (phospho T71) antibody [E268] (ab32019)



Western blot - Anti-ATF2 (phospho T71) antibody [E268] (ab32019)

All lanes : Anti-ATF2 (phospho T71) antibody [E268] (ab32019) at 0.1 µg/ml

Lane 1 : HeLa (human cervix adenocarcinoma), prepared in FBS free medium overnight, whole cell lysate

Lane 2: HeLa, prepared in FBS free medium overnight, then treated with 250ng/ml anisomycin for 30 minutes whole cell lysate

Lysates/proteins at 10 µg per lane.

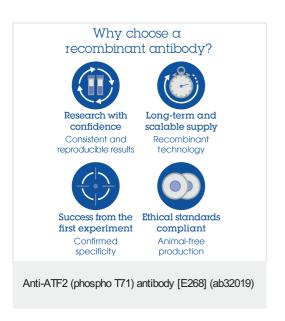
Secondary

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

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

Exposure time: 10 seconds

Blocking and diluting buffer used was 2% BSA/TBST . The molecular weight is 70kD, consistent with the literature (PMID: 24223142).



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