abcam

Product datasheet

Anti-c-Fos (phospho T325) antibody ab27793

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製品の概要

製品名	Anti-c-Fos (phospho T325) antibody
製品の詳細	Rabbit polyclonal to c-Fos (phospho T325)
由来種	Rabbit
特異性	ab27793 recognises the cFos phosphorilated at threonine 325 form.
アプリケーション	**適用あり:** ICC/IF, ChIP, WB
種交差性	**交差種:** Human
免疫原	Synthetic peptide corresponding to Human c-Fos (phospho T325).
ポジティブ・コントロール	WB: A431 cell lysate. ICC: HeLa cells. ChIP: A431 cells.
特記事項	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. 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, along with publications, customer reviews and Q&As

製品の特性

製品の状態	Liquid
保存方法	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term.
バッファー	pH: 7.30 Preservative: 0.05% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine), 0.1% BSA
精製度	Immunogen affinity purified
特記事項(精製)	ab27793 was negatively preadsorbed using a non phosphopeptide corresponding to the site of phosphorylation to remove antibody that is reactive with non phosphorylated cFos. Immunogen affinity purification followed.
ポリ/モノ	ポリクローナル
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ab27793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
ICC/IF		1/250.
ChIP		Use 1-3µg for 10^6 cells.
WB		1/1000. Predicted molecular weight: 41 kDa.

ターゲット情報

機能

Nuclear phosphoprotein which forms a tight but non-covalently linked complex with the JUN/AP-1 transcription factor. In the heterodimer, FOS and JUN/AP-1 basic regions each seems to interact with symmetrical DNA half sites. On TGF-beta activation, forms a multimeric SMAD3/SMAD4/JUN/FOS complex at the AP1/SMAD-binding site to regulate TGF-beta-mediated signaling. Has a critical function in regulating the development of cells destined to form and maintain the skeleton. It is thought to have an important role in signal transduction, cell proliferation and differentiation.

配列類似性

Belongs to the bZIP family. Fos subfamily.
Contains 1 bZIP domain.

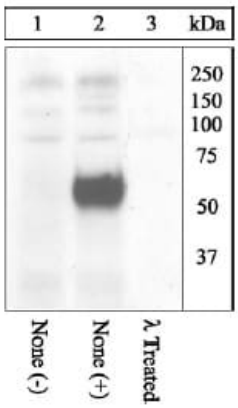
翻訳後修飾

Phosphorylated in the C-terminal upon stimulation by nerve growth factor (NGF) and epidermal growth factor (EGF). Phosphorylated, in vitro, by MAPK and RSK1. Phosphorylation on both Ser-362 and Ser-374 by MAPK1/2 and RSK1/2 leads to protein stabilization with phosphorylation on Ser-374 being the major site for protein stabilization on NGF stimulation. Phosphorylation on Ser-362 and Ser-374 primes further phosphorylations on Thr-325 and Thr-331 through promoting docking of MAPK to the DEF domain. Phosphorylation on Thr-232, induced by HA-RAS, activates the transcriptional activity and antagonizes sumoylation. Phosphorylation on Ser-362 by RSK2 in osteoblasts contributes to osteoblast transformation.
Constitutively sumoylated by SUMO1, SUMO2 and SUMO3. Desumoylated by SENP2. Sumoylation requires heterodimerization with JUN and is enhanced by mitogen stimulation. Sumoylation inhibits the AP-1 transcriptional activity and is, itself, inhibited by Ras-activated phosphorylation on Thr-232.

細胞内局在

Nucleus.

画像



Western blot - Anti-c-Fos (phospho T325) antibody (ab27793)

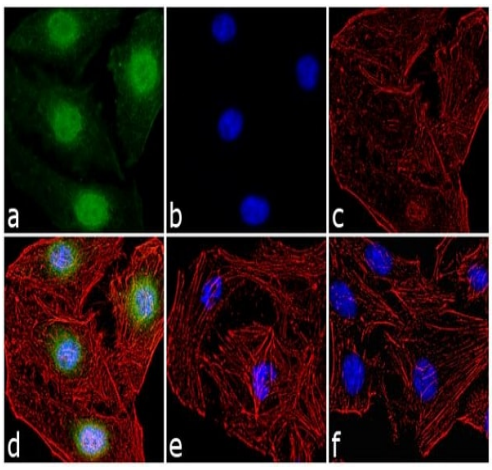
All lanes : Anti-c-Fos (phospho T325) antibody (ab27793) at 1/1000 dilution (diluted in a 3% Milk TBST buffer.)

Lane 1 : Non EGF treated A431 cell lysate
Lane 2 : EGF treated A431 cell lysate
Lane 3 : EGF treated A431 cell lysate treated with Lambda phosphatase

Secondary
All lanes : goat F(ab')2 anti rabbit IgG HRP conjugate

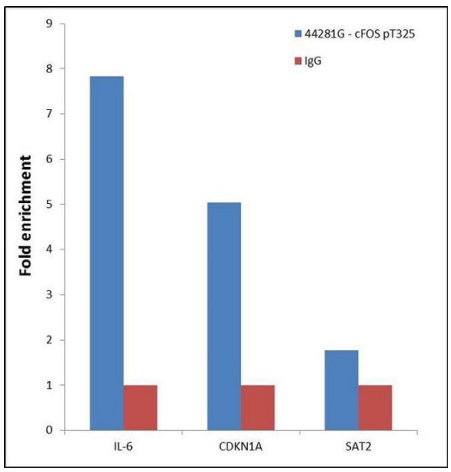
Predicted band size: 41 kDa
Observed band size: 58 kDa

The figure shows that the phosphorylation of cFos on threonine 325 is induced by EGF treatment and that Lambda phosphatase treatment eliminates the signal, thereby demonstrating the phospho specificity of ab27793.



Immunocytochemistry/ Immunofluorescence - Anti-c-Fos (phospho T325) antibody (ab27793)

Immunofluorescence analysis of c-Fos (phospho T325) was done on 70% confluent log phase HeLa cell treated with 200 nM of PMA for 20 minutes. The cell were fixed with 4% paraformaldehyde for 10 minutes, permeabilized with 0.1% Triton™ X-100 for 10 minutes, and blocked with 1% BSA for 1 hour at room temperature. The cells were labeled with Phosc-Fos (phospho T325)Rabbit Polyclonal Antibody (ab27793) at 2 ug/ml in 0.1% BSA and incubated for 3 hours at room temperature and then labeled with Goat anti-Rabbit IgG (H+L) Secondary Antibody, Alexa Fluor® 488 conjugate at a dilution of 1/2000 for 45 minutes at room temperature (Panel a: green). Nuclei (Panel b: blue) were stained with DAPI. F-actin (Panel c: red) was stained with Alexa Fluor® 555 Rhodamine Phalloidin. Panel d is a merged image showing nuclear and cytoplasmic localization. Panel e is untreated cell with no signal. Panel f represents control cells with no primary antibody to assess background. The images were captured at 60X magnification.



ChIP - Anti-c-Fos (phospho T325) antibody (ab27793)

Chromatin Immunoprecipitation (ChIP) was performed using Anti-c-Fos (phospho T325) antibody (ab27793) 3 ug on sheared chromatin from 2 million A431 cells treated with EGF (200ng/ml), for 30 minutes. Normal Rabbit IgG was used as a negative IP control. The purified DNA was analyzed by 7500 Fast qPCR system with optimized PCR primer pairs for the promoter of active IL-6, CDKN1A gene, used as positive control target, and the SAT2, used as negative control target. Data is presented as fold enrichment of the antibody signal versus the negative control IgG using the comparative CT method.

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