abcam

Product datasheet

Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody ab4821

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概述

产品名称	Anti-JNK1 + JNK2 (phospho T183 + Y185)**抗体**
描述	**兔多克隆抗体**to JNK1 + JNK2 (phospho T183 + Y185)
宿主	Rabbit
特异性	Phosphorylation site-specific antibody selective for the dually phosphorylated form of the c-Jun N-terminal Kinase (JNK)/Stress-Activated Protein Kinase (SAPK) enzymes containing a phosphate on threonine 183 and tyrosine 185 (human JNK 1 + 2). The antibody has been shown to recognize the endogenous, active forms of JNK 1 + 2 in a variety of cell types following treatment by a broad range of extracellular stimuli [e.g. including 293 cells (human embryonic kidney; +/- ultraviolet light) and PC12 cells (rat pheochromocytoma; +/- sorbital)]. The region of JNK1 and JNK2 surrounding T183 + Y185 has a high degree of similarity to the corresponding regions in JNK3 and thus may cross react with this protein if phosphorylated on the corresponding residues.
经测试应用	**适用于:** ICC/IF, WB
种属反应性	**与反应:** Mouse, Human **预测可用于:** a wide range of other species
免疫原	Synthetic peptide corresponding to Human JNK1 + JNK2 (phospho T183 + Y185).
常规说明	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

性能

形式	Liquid
存放说明	Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.
存储溶液	pH: 7.30 Preservative: 0.05% Sodium azide Constituents: PBS, 50% Glycerol, 0.1% BSA

	BSA is IgG and protease free
纯度	Immunogen affinity purified
纯化说明	Purified from rabbit serum by sequential epitope specific chromatography. The antibody has been negatively preadsorbed using a non-phosphopeptide corresponding to the site of phosphorylation to remove antibody that is reactive with non-phosphorylated JNK enzymes. The final product is generated by affinity chromatography using a JNK-derived peptide that is phosphorylated at threonine 183 and tyrosine 185, within the activation loop. Note: It is the dually phosphorylated form of these enzymes that has full enzymatic activity.
克隆	多克隆
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab4821于以下的经测试应用

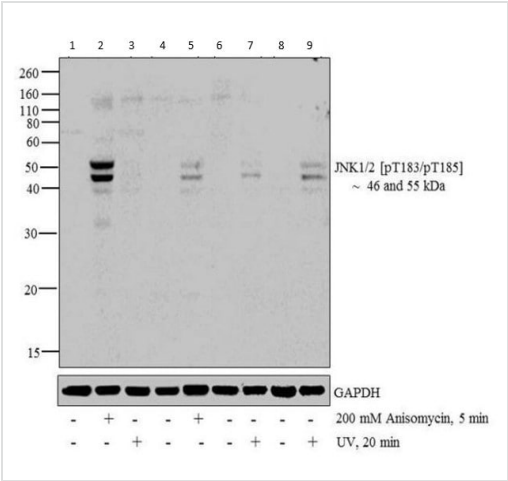
"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ICC/IF	★★★★☆ (1)	1/250. 1/100.
WB	★★★★☆ (7)	1/1000. Predicted molecular weight: 49, 55 kDa. Band at ~49 kDa represents Jnk1, while the band at ~55 kDa represents Jnk2

靶标

功能	Responds to activation by environmental stress and pro-inflammatory cytokines by phosphorylating a number of transcription factors, primarily components of AP-1 such as JUN, JDP2 and ATF2 and thus regulates AP-1 transcriptional activity. In T-cells, JNK1 and JNK2 are required for polarized differentiation of T-helper cells into Th1 cells (By similarity). Phosphorylates heat shock factor protein 4 (HSF4). JNK1 isoforms display different binding patterns: beta-1 preferentially binds to c-Jun, whereas alpha-1, alpha-2, and beta-2 have a similar low level of binding to both c-Jun or ATF2. However, there is no correlation between binding and phosphorylation, which is achieved at about the same efficiency by all isoforms.
序列相似性	Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. MAP kinase subfamily. Contains 1 protein kinase domain.
结构域	The TXY motif contains the threonine and tyrosine residues whose phosphorylation activates the MAP kinases.
翻译后修饰	Dually phosphorylated on Thr-183 and Tyr-185, which activates the enzyme.

图片



Western blot - Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821)

All lanes : Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821) at 1/1000 dilution

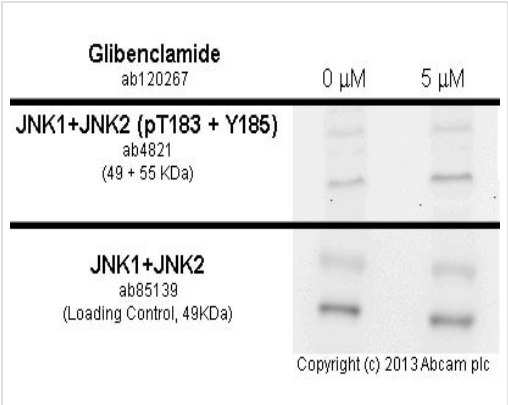
Lane 1 : HEK-293 cell line
Lane 2 : HEK-293 treated for 5 minutes with 200 mM of Anisomycin
Lane 3 : HEK-293 treated for 20 minutes with UV
Lane 4 : MCF7 cell line
Lane 5 : MCF7 treated for 5 minutes with 200 mM of Anisomycin
Lane 6 : K562 cell line
Lane 7 : K562 treated for 20 minutes with UV
Lane 8 : HeLa cell line
Lane 9 : HeLa treated for 20 minutes with UV

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG - HRP Secondary Antibody at 1/5000 dilution

Predicted band size: 49, 55 kDa

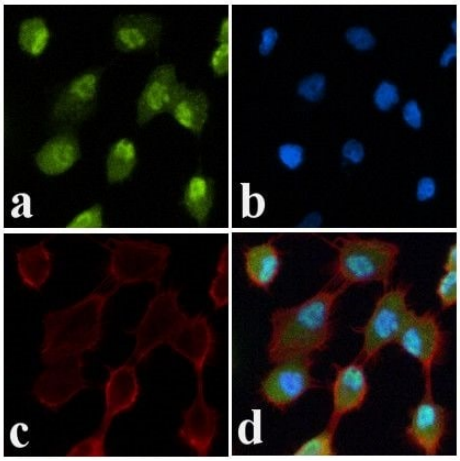
Proteins were transferred to a nitrocellulose membrane and blocked with 5% skim milk for 1 hour at room temperature.



Western blot - Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821)

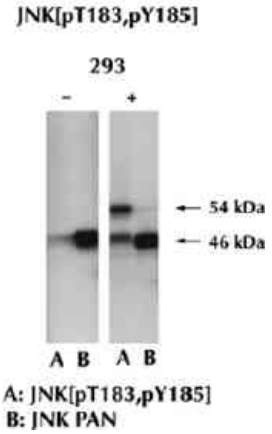
MEF1 cells were incubated at 37°C for 48h with vehicle control (0 µM) and 5 µM of glibenclamide (**ab120267**) in DMSO. Increased expression of of JNK1+JNK2 (phospho T183 + Y185) (ab4821) correlates with an increase in glibenclamide concentration, as described in literature.

Whole cell lysates were prepared with RIPA buffer (containing protease inhibitors and sodium orthovanadate), 10µg of each were loaded on the gel and the WB was run under reducing conditions. After transfer the membrane was blocked for an hour using 3% milk before being incubated with ab4821 at 1/1000 dilution and **ab85139** at 1 µg /ml overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP (**ab97051**) at 1/10000 dilution and visualised using ECL development solution.



Immunocytochemistry/ Immunofluorescence - Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821)

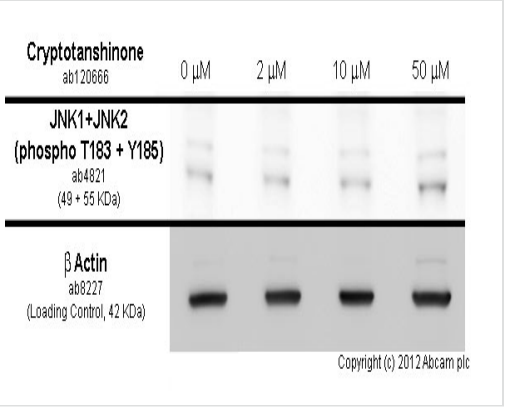
ab4821 staining JNK1 + JNK2 (phospho T183 + Y185) in A549 cells (green, panel a) by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with 4% paraformaldehyde, permeabilized with 0.25% Triton X-100 and blocked with 5% BSA for 1 hour at room temperature. Samples were incubated with primary antibody (2ug/ml in 1% BSA) for 3 hours at room temperature. An Alexa Fluor® 488-conjugated Goat anti-rabbit IgG polyclonal was used as the secondary antibody (1/400). Nuclei stained with DAPI (blue, panel b), F-actin stained with Alexa Fluor® 594 Phalloidin (red, panel b) and merged images (panel d).



Western blot - Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821)

To demonstrate the phosphorylation of JNK 1 & 2 in a cell based assay, 293 cells were treated with ultraviolet irradiation (UV). Proteins from cell extracts were resolved by SDS-PAGE on a 10% Tris-glycine gel and transferred to nitrocellulose. Membranes were incubated with either 1 µ g/mL ab4821 or 1 µg/mL anti-JNK1 pan. After washing, membranes were incubated with goat F(ab')2 anti-rabbit IgG alkaline phosphatase and bands were detected using the Tropix WesternStar detection method.

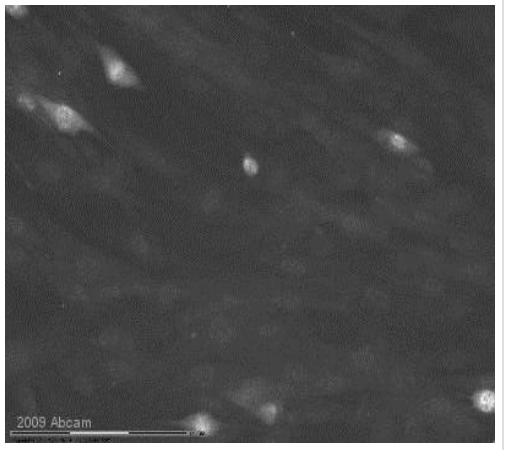
To demonstrate the phosphorylation of JNK 1 & 2 in a cell based assay, 293 cells were treated with ultraviolet irradiation (UV). Proteins from cell extracts were resolved by SDS-PAGE on a 10% Tris-glycine gel and transferred to nitrocellulose. Membranes were incubated with either 1 µ g/mL ab4821 or 1 µg/mL anti-JNK1 pan. After washing, membranes were incubated with goat F(ab')2 anti-rabbit IgG alkaline phosphatase and bands were detected using the Tropix We



Western blot - Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821)

MCF7cells were incubated at 37°C for 4h with vehicle control (0 µM) and different concentrations of cryptotanshinone (**ab120666**). Increased expression of JNK1+JNK2 (phospho T183 + Y185) in MCF7 cells correlates with an increase in cryptotanshinone concentration, as described in literature.

Whole cell lysates were prepared with RIPA buffer (containing protease inhibitors and sodium orthovanadate), 10µg of each were loaded on the gel and the WB was run under reducing conditions. After transfer the membrane was blocked for an hour using 5% BSA before being incubated with ab4821 at 1/1000 dilution and **ab8227** at 1 µg/ml overnight at 4°C. Antibody binding was detected using an anti-rabbit antibody conjugated to HRP (**ab97051**) at 1/10000 dilution and visualised using ECL development solution.



Immunocytochemistry/ Immunofluorescence - Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821)

This image is courtesy of an Abreview submitted by Mr George Chennell

ab4821 staining JNK1+JNK2 (phospho T183 + Y185) in human foreskin fibroblasts by ICC/IF. The cells were fixed in cytoskeletal fixative, permeabilized in 0.5% Triton X-100 and blocked in 2% dillution buffer (2%BSA + 0.1% Triton X-100) for 1 hour at 25°C. The primary antibody was diluted, 1/100 and incubated with sample for 12 hours. An Alexa Fluor® 594 conjugated goat polyclonal to rabbit IgG, diluted 1/250 was used as secondary.

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