


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

★★★★★ **9 Abreviews** **90 References** 画像数 6

#### 製品の概要

製品名	Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody
製品の詳細	Rabbit polyclonal to JNK1 + JNK2 (phospho T183 + Y185)
由来種	Rabbit
特異性	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.
アプリケーション	<b>適用あり:</b> ICC/IF, WB
種交差性	<b>交差種:</b> Mouse, Human <b>交差が予測される動物種:</b> a wide range of other species 
免疫原	Synthetic peptide corresponding to Human JNK1 + JNK2 (phospho T183 + Y185).
特記事項	<p>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.</p> <p>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&amp;As</p>

#### 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 or -80°C. Avoid repeated freeze / thaw cycles.
バッファー	pH: 7.30 Preservative: 0.05% Sodium azide Constituents: PBS, 50% Glycerol, 0.1% BSA

	BSA is IgG and protease free
精製度	Immunogen affinity purified
特記事項 (精製)	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.
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

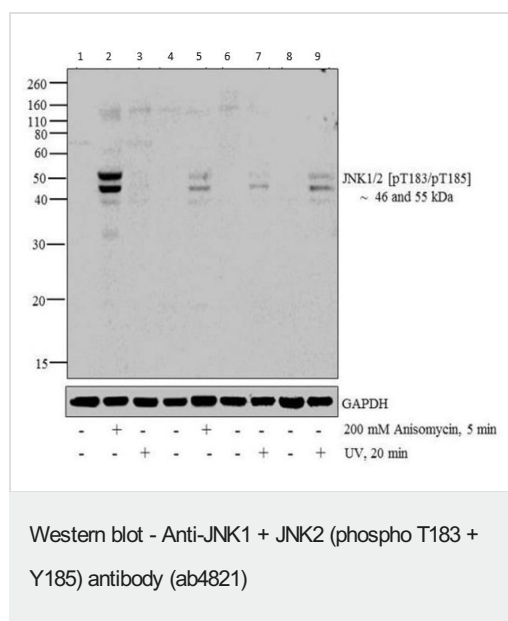
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アプリケーション	Abreviews	特記事項
ICC/IF	★★★★★ (1)	1/250. 1/100.
WB	★★★★★ (7)	1/1000. Predicted molecular weight: 49, 55 kDa. Band at ~49 kDa represents Jnk1, while the band at ~55 kDa represents Jnk2

## ターゲット情報

機能	<p>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).</p> <p>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.</p>
配列類似性	<p>Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. MAP kinase subfamily.</p> <p>Contains 1 protein kinase domain.</p>
ドメイン	The TXY motif contains the threonine and tyrosine residues whose phosphorylation activates the MAP kinases.
翻訳後修飾	Dually phosphorylated on Thr-183 and Tyr-185, which activates the enzyme.

## 画像



**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

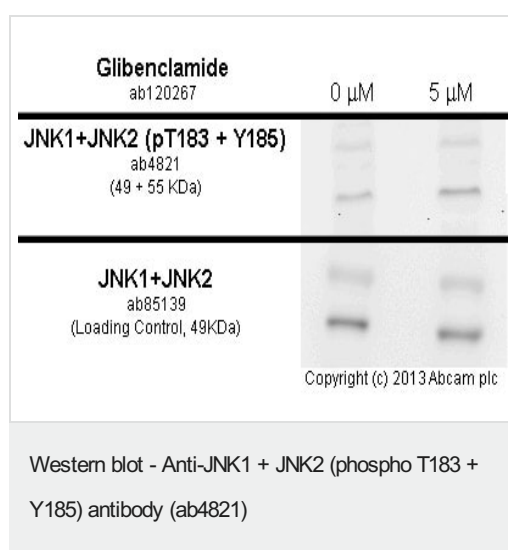
Lysates/proteins at 20 µg per lane.

## Secondary

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

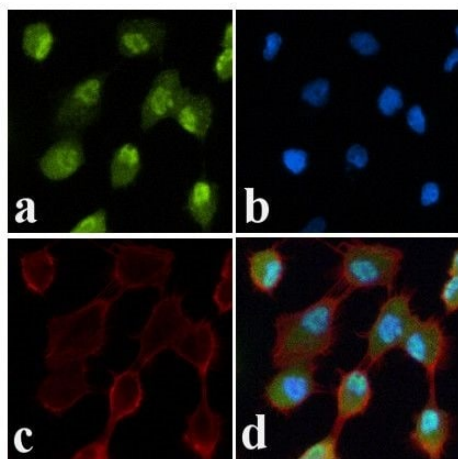
**Predicted band size:** 49, 55 kDa

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

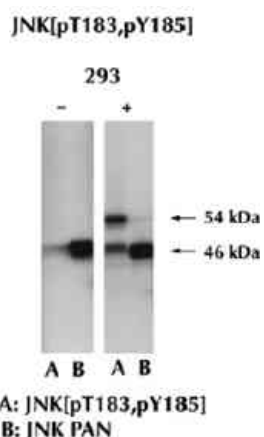


MEF1 cells were incubated at 37°C for 48h with vehicle control (0 µM) and 5 µM of glibenclamide (**ab120267**) in DMSO. Increased expression of JNK1+JNK2 (phospho T183 + Y185) (ab4821) correlates with an increase in glibenclamide concentration, as described in literature.

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)

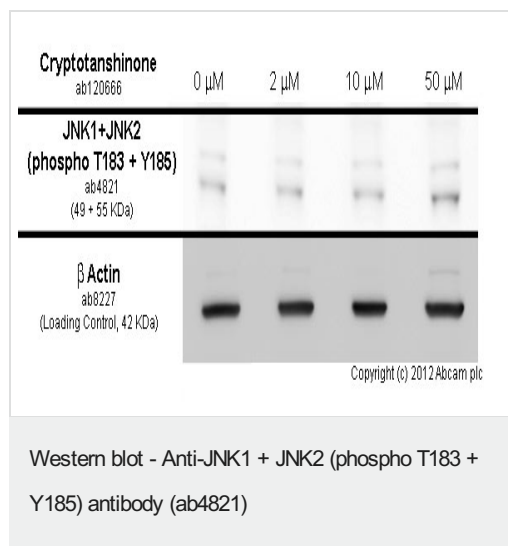


Western blot - Anti-JNK1 + JNK2 (phospho T183 + Y185) antibody (ab4821)

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 c) and merged images (panel d).

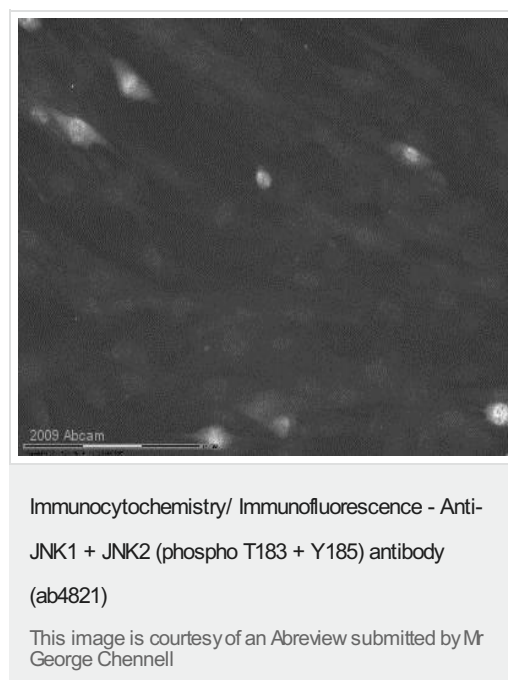
To demonstrate the phosphorylation of JNK 1 & 2 in a cell based assay, 293 cells were treated with ultraviolet irradiation (UV). Proteins from cell extracts were resolved by SDS-PAGE on a 10% Tris-glycine gel and transferred to nitrocellulose. Membranes were incubated with either 1 µg/mL ab4821 or 1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



MCF7 cells 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.



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% dilution 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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