abcam

Product datasheet

# Anti-c-Jun (phospho S63) antibody [Y172] ab32385

リコンビナント RabMAb

4 Abreviews 78 References 画像数 11

## 製品の概要

| | |
|---|---|
| **製品名** | Anti-c-Jun (phospho S63) antibody [Y172] |
| **製品の詳細** | Rabbit monoclonal [Y172] to c-Jun (phospho S63) |
| **由来種** | Rabbit |
| **特異性** | Anti-c-Jun (phospho S63) antibody [Y172] (ab32385) only detects c-Jun phosphorylated on Serine 63 when tested in WB and ICC using specific phospho-treatments. However, in DotBlot and ELISA assays we detected some cross-reactivity with the non-phospho peptide as well. Please refer to the images on the datasheet. The mouse recommendation is based on the WB results. We do not guarantee IHC-P for mouse. |
| **アプリケーション** | **適用あり:** WB, IHC-P, ICC/IF, Dot blot, ELISA<br>**適用なし:** Flow Cyt |
| **種交差性** | **交差種:** Mouse, Human<br>**交差が予測される動物種:** Rat, Cow |
| **免疫原** | Synthetic peptide within Human c-Jun aa 50-150 (phospho S63). The exact sequence is proprietary.<br>Database link: **P05412** |
| **ポジティブ・コントロール** | WB: UV or Anisomycin treated NIH/3T3 or HeLa whole cell lysate (**ab150035**). IHC-P: Human breast carcinoma tissue. ICC/IF: A431 cells, NIH/3T3 cells. |
| **特記事項** | This product is a recombinant monoclonal antibody, which offers several advantages including:<br>- High batch-to-batch consistency and reproducibility<br>- Improved sensitivity and specificity<br>- Long-term security of supply<br>- Animal-free production<br>For more information **see here**.<br>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. |

## 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. Avoid freeze / thaw cycle. |
| **バッファー** | pH: 7.20<br>Preservative: 0.01% Sodium azide<br>Constituents: 59% PBS, 40% Glycerol, 0.05% BSA |
| **精製度** | Protein A purified |
| **ポリ/モノ** | モノクローナル |
| **クローン名** | Y172 |
| **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ab32385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** | ★★★★★ (2) | 1/1000 - 1/10000. Detects a band of approximately 42 kDa (predicted molecular weight: 36 kDa). |
| **IHC-P** | | 1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.<br>**For unpurified use at 1/50 - 1/100**<br>The mouse recommendation is based on the WB results. We do not guarantee IHC-P for mouse. |
| **ICC/IF** | | 1/100 - 1/200. |
| **Dot blot** | | 1/1000. |
| **ELISA** | | Use at an assay dependent concentration. |

**追加情報** Is unsuitable for Flow Cyt.

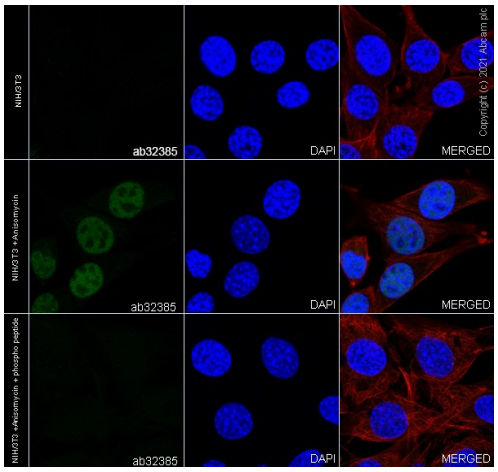
## ターゲット情報

| | |
|---|---|
| **機能** | Transcription factor that recognizes and binds to the enhancer heptamer motif 5'-TGA[CG]TCA-3'. Promotes activity of NR5A1 when phosphorylated by HIPK3 leading to increased steroidogenic gene expression upon cAMP signaling pathway stimulation. Involved in activated KRAS-mediated transcriptional activation of USP28 in colorectal cancer (CRC) cells (PubMed:24623306). Binds to the USP28 promoter in colorectal cancer (CRC) cells (PubMed:24623306). |
| **配列類似性** | Belongs to the bZIP family. Jun subfamily.<br>Contains 1 bZIP (basic-leucine zipper) domain. |
| **翻訳後修飾** | Ubiquitinated by the SCF(FBXW7), leading to its degradation. Ubiquitination takes place following phosphorylation, that promotes interaction with FBXW7. |

Phosphorylated by CaMK4 and PRKDC; phosphorylation enhances the transcriptional activity. Phosphorylated by HIPK3. Phosphorylated by DYRK2 at Ser-243; this primes the protein for subsequent phosphorylation by GSK3B at Thr-239. Phosphorylated at Thr-239, Ser-243 and Ser-249 by GSK3B; phosphorylation reduces its ability to bind DNA. Phosphorylated by PAK2 at Thr-2, Thr-8, Thr-89, Thr-93 and Thr-286 thereby promoting JUN-mediated cell proliferation and transformation. Phosphorylated by PLK3 following hypoxia or UV irradiation, leading to increase DNA-binding activity.
Acetylated at Lys-271 by EP300.

**細胞内局在** Nucleus.

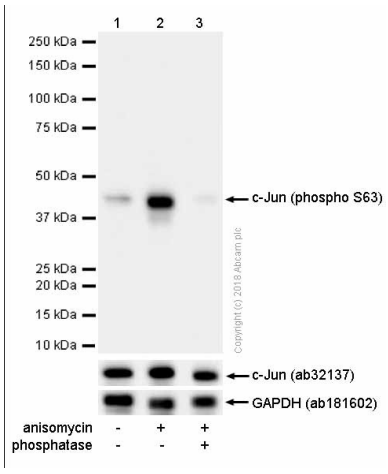
## 画像




Immunocytochemistry/ Immunofluorescence - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

Immunocytochemistry confocal image of 4% paraformaldehyde-fixed 0.1% Triton X-100 permeabilized anisomycin-treated NIH/3T3 cell line (mouse embryonic fibroblast), staining nuclear c-Jun with ab32385 at 1:500 dilution and **ab150077** AlexaFluor®488 Goat anti-Rabbit secondary at 1:1000 dilution. The counterstain was **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1:200 dilution, and the nuclear counterstain was DAPI (blue).

The NIH/3T3 cells were treated with 250 ng/ml Anisomycin for 30 minutes and then the signal decreased after phosphatase treatment at 37°C for 2 hours.




Western blot - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

**All lanes :** Anti-c-Jun (phospho S63) antibody [Y172] (ab32385) at 0.1 µg/ml (purified)

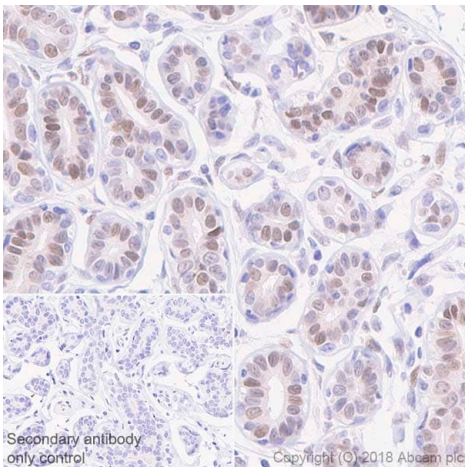
**Lane 1 :** NIH/3T3 (Mouse embryonic fibroblast) whole cell lysates
**Lane 2 :** NIH/3T3 (Mouse embryonic fibroblast) treated with 250 ng/ml anisomycin for 30 minutes whole cell lysates
**Lane 3 :** NIH/3T3 (Mouse embryonic fibroblast) treated with 250 ng/ml anisomycin for 30 minutes whole cell lysates. Then the membrane was incubated with phosphatase

Lysates/proteins at 15 µg per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

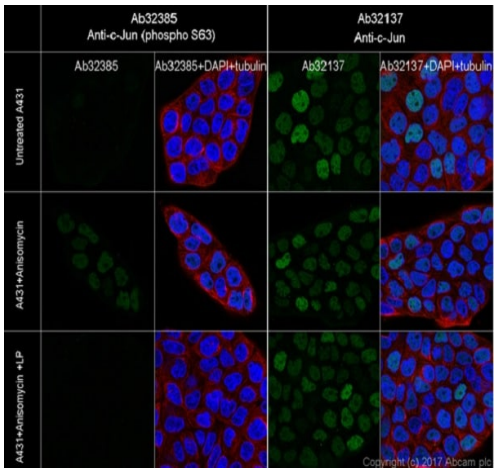
**Predicted band size:** 36 kDa

Blocking and diluting buffer: 5% NFDM/TBST.



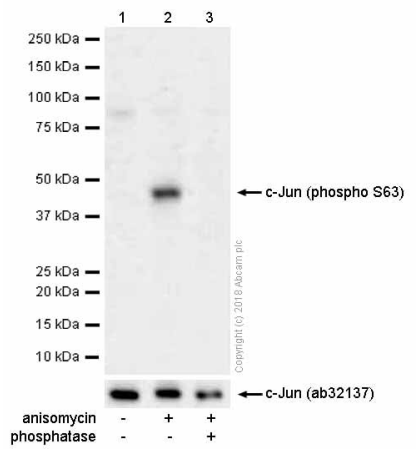

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast tissue sections labeling c-Jun with Purified ab32385 at 1:250 dilution (0.46 µg/ml). Heat mediated antigen retrieval was performed using using **ab93684** (Tris/EDTA buffer, pH 9.0)ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody.Negative control:PBS instead of the primary antibody.Hematoxylinwas used as a counterstain




Immunocytochemistry/ Immunofluorescence - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized A431 (Human epidermoid carcinoma cell line) cells labeling c-Jun (phospho S63) with ab32385 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing the expression was increased after treatment with anisomycin (1 µg/ml for 15 minutes), then decreased after treatment with the Lambda Protein Phosphatase treatment 31 for 2 hours. The nuclear counter stain is DAPI (blue). Counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at a 1/200 dilution (red).




Western blot - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

**All lanes :** Anti-c-Jun (phospho S63) antibody [Y172] (ab32385) at 0.1 µg/ml (purified)

**Lane 1 :** HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates
**Lane 2 :** HeLa (Human cervix adenocarcinoma epithelial cell) treated with 1 ug/ml anisomycin for 15 minutes whole cell lysates
**Lane 3 :** HeLa (Human cervix adenocarcinoma epithelial cell) treated with 1 ug/ml anisomycin for 15 minutes whole cell lysates 15ug. Then the membrane was incubated with phosphatase.

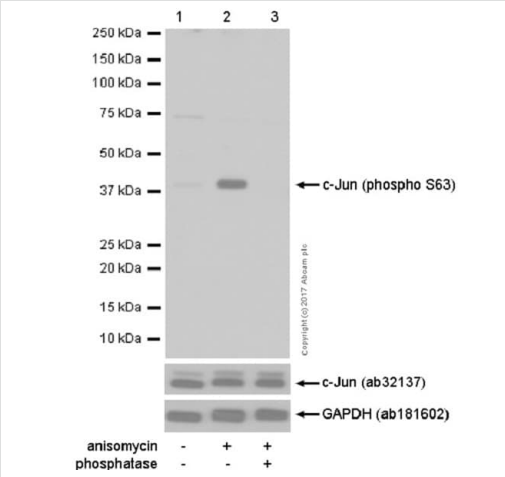
Lysates/proteins at 15 µg per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

**Predicted band size:** 36 kDa

Blocking and diluting buffer: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab32385).




Western blot - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

**Lane 1 :** Anti-c-Jun (phospho S63) antibody [Y172] (ab32385) at 1/1000 dilution (Unpurified)
**Lanes 2-3 :** Human HRPT2/Parafibromin peptide (**ab23385**) at 1/1000 dilution (Unpurified)

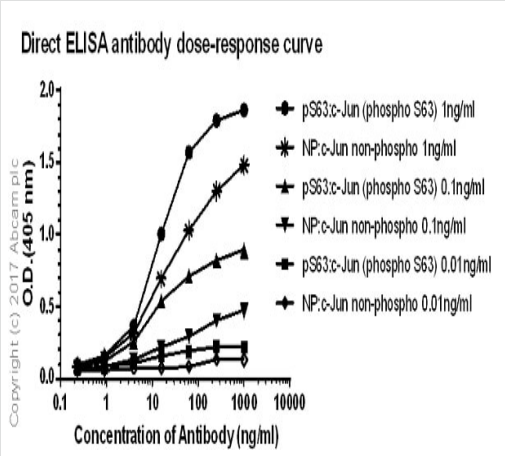
**Lane 1 :** HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates with NFDM/TBST
**Lane 2 :** HeLa (Human cervix adenocarcinoma epithelial cell) treated with 1ug/mL anisomycin for 15 minutes whole cell lysates with NFDM/TBST
**Lane 3 :** HeLa (Human cervix adenocarcinoma epithelial cell) treated with 1ug/ml anisomycin for 15 minutes whole cell lysates. Then the membrane was incubated with phosphatase. with NFDM/TBST

Lysates/proteins at 15 µg per lane.

Blocking peptides at 5 % per lane.

**Secondary**
**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

**Predicted band size:** 36 kDa

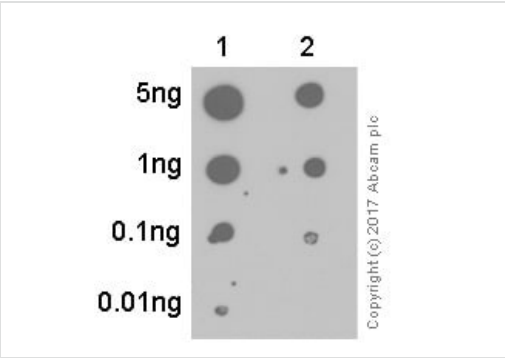



ELISA - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

Antigen pS63:c-Jun (phospho S63); NP:c-Jun non-phospho. Antigen concentration 0.01~1 ng/ml.

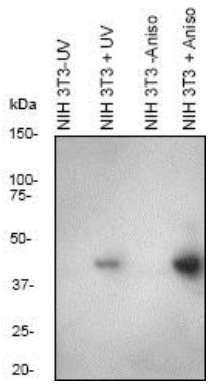
Primary antibody concentration range 0~1000 ng/ml.

Secondary antibody is an Alkaline Phosphatase-conjugated Goat Anti-Rabbit IgG(H+L) used at a 1:2500 dilution.




Dot Blot - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

Unpurified ab32385 used at a 1:1000 dilution.
Secondary antibody is Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (**ab97051**) used at a 1:100,000 dilution.
Blocking/Diluting buffer and concentration: 5% NFDM/TBST.
**Lane 1:** Human c-Jun (pS63) phospho peptide.
**Lane 2:** Human c-Jun non-phospho peptide.
Exposure time 3 minutes.

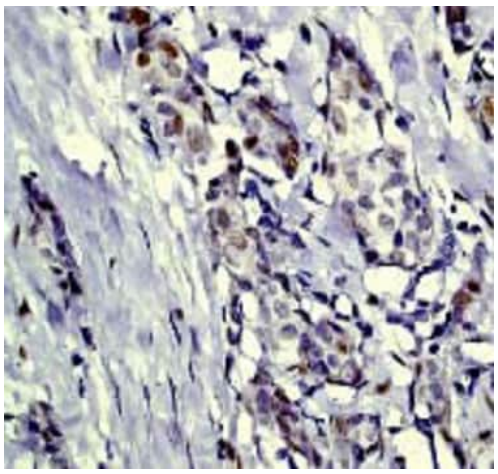



Western blot - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

**All lanes :** Anti-c-Jun (phospho S63) antibody [Y172] (ab32385) at 1/10000 dilution (Unpurified)

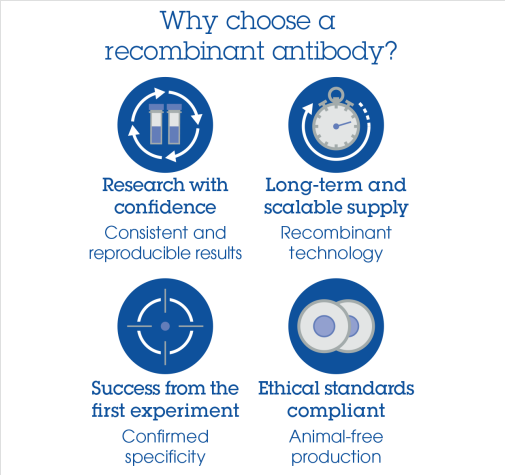
**Lanes 1 & 3 :** Untreated NIH/3T3 (Mouse embyro fibroblast cell line) cell lysate
**Lane 2 :** NIH/3T3 (Mouse embyro fibroblast cell line) cell lysate treated with ultraviolet light
**Lane 4 :** NIH/3T3 (Mouse embyro fibroblast cell line) cell lysate treated with 25 µg/ml Anisomycin for 15 minutes at 37°C

**Predicted band size:** 36 kDa
**Observed band size:** 42 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

Paraffin-embedded human breast carcinoma tissue stained for c-Jun (phospho S63) with unpurified ab32385 at a 1/50 dilution in immunohistochemical analysis.




Anti-c-Jun (phospho S63) antibody [Y172] (ab32385)

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