

Anti-Phospho-c-Jun (S63) Antibody [SY0297] - BSA and Azide free

HA750146



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 36 kDa
Clone number:	SY0297

Description: c-Jun is a protein that in humans is encoded by the JUN gene. c-Jun, in combination with c-Fos, forms the AP-1 early response transcription factor. It was first identified as the Fos-binding protein p39 and only later rediscovered as the product of the JUN gene. c-jun was the first oncogenic transcription factor discovered. The proto-oncogene c-Jun is the cellular homolog of the viral oncoprotein v-jun (P05411). The viral homolog v-jun was discovered in avian sarcoma virus 17 and was named for ju-nana, the Japanese word for 17. The human JUN encodes a protein that is highly similar to the viral protein, which interacts directly with specific target DNA sequences to regulate gene expression. This gene is intronless and is mapped to 1p32-p31, a chromosomal region involved in both translocations and deletions in human malignancies.

Immunogen: Synthetic phospho-peptide corresponding to residues surrounding Ser63 of Human c-Jun aa 31-80 / 331.

Positive control: C6 treated with 25µg/mL Anisomycin for 30 minutes cell lysate, A549 treated with 250ng/mL anisomycin for 30 minutes whole cell lysate, NIH/3T3 treated with 250ng/mL anisomycin for 30 minutes whole cell lysate, PC-3M, MCF-7, A549, human breast carcinoma tissue, human tonsil tissue, human colon carcinoma tissue, human endometrial tissue.

Subcellular location: Nucleus.

Database links: SwissProt: P05412 Human | P05627 Mouse | P17325 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:50-1:200
IHC-P	1:50-1:500

Storage Buffer: 1*PBS (pH7.4).

Storage Instruction: Store at +4°C after thawing. Aliquot store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

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Orders:0086-571-88062880

Technical:0086-571-89986345

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Images

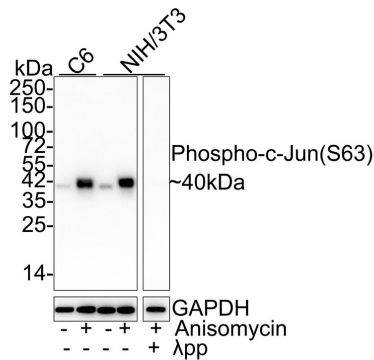


Fig1: Western blot analysis of Phospho-c-Jun (S63) on different lysates with Rabbit anti-Phospho-c-Jun (S63) antibody (HA750146) at 1/2,000 dilution.

Lane 1: C6 cell lysate

Lane 2: C6 treated with 25μg/mL Anisomycin for 30 minutes cell lysate

Lane 3: NIH/3T3 cell lysate

Lane 4: NIH/3T3 treated with 25μg/mL Anisomycin for 30 minutes cell lysate

Lane 5: NIH/3T3 treated with 25μg/mL Anisomycin for 30 minutes, then treated with λpp for 1 hour cell lysate

Lysates/proteins at 20 μg/Lane.

Predicted band size: 36 kDa

Observed band size: 40 kDa

Exposure time: 1 minute;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDm/TBST for 1 hour at room temperature. The primary antibody (HA750146) at 1/2,000 dilution was used in 5% NFDm/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

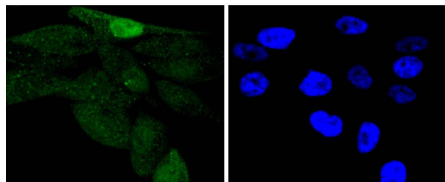


Fig2: ICC staining of Phospho-c-Jun (S63) in PC-3M cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750146, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor@488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

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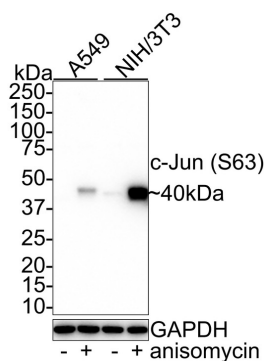
Fig3: Western blot analysis of Phospho-c-Jun (S63) on different lysates with Rabbit anti-Phospho-c-Jun (S63) antibody (HA750146) at 1/500 dilution.

Lane 1: A549 whole cell lysate

Lane 2: A549 treated with 250ng/mL anisomycin for 30 minutes whole cell lysate

Lane 3: NIH/3T3 whole cell lysate

Lane 4: NIH/3T3 treated with 250ng/mL anisomycin for 30 minutes whole cell lysate



Lysates/proteins at 20 µg/Lane.

Predicted band size: 36 kDa

Observed band size: 40 kDa

Exposure time: 5 minutes;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750146) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:100,000 dilution was used for 1 hour at room temperature.

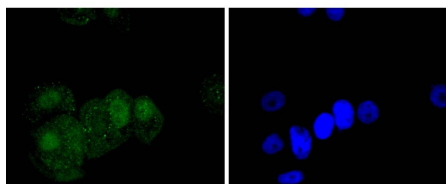


Fig4: ICC staining of Phospho-c-Jun (S63) in MCF-7 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750146, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor@488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

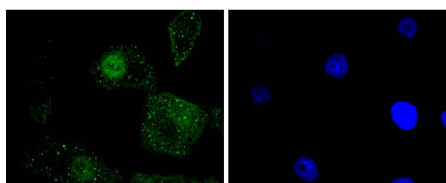


Fig5: ICC staining of Phospho-c-Jun (S63) in A549 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750146, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor@488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

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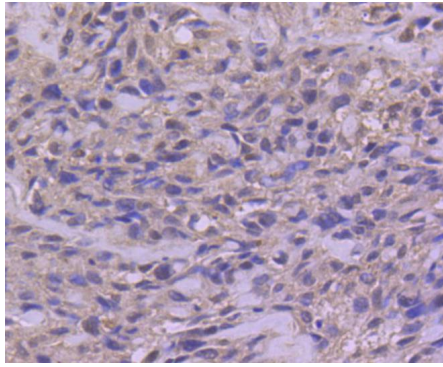


Fig6: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue using anti-Phospho-c-Jun (S63) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750146, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

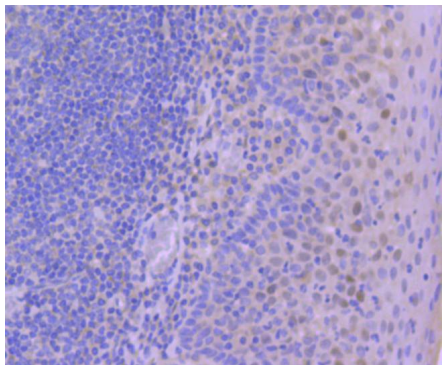


Fig7: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-Phospho-c-Jun (S63) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750146, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

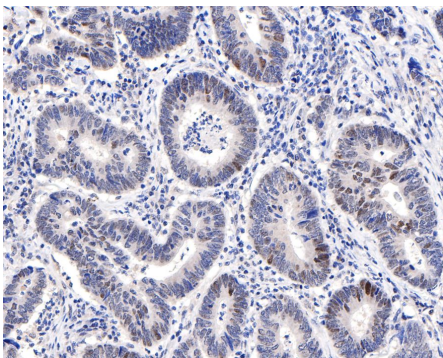


Fig8: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue with Rabbit anti-Phospho-c-Jun (S63) antibody (HA750146) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750146) at 1/500 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

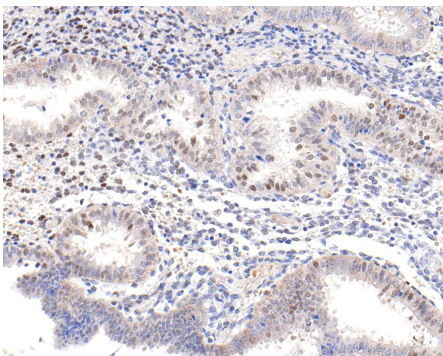


Fig9: Immunohistochemical analysis of paraffin-embedded human endometrial tissue with Rabbit anti-Phospho-c-Jun (S63) antibody (HA750146) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750146) at 1/500 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Zhang QS et al. Downregulation of SENP1 inhibits cell proliferation, migration and promotes apoptosis in human glioma cells. *Oncol Lett* 12:217-221 (2016).
2. Li C et al. Inhibitory effects of kaempferol on the invasion of human breast carcinoma cells by downregulating the expression and activity of matrix metalloproteinase-9. *Biochem Cell Biol* 93:16-27 (2015).

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