

Anti-p38 alpha / MAPK14 Antibody [SR43-04] - BSA and Azide free

HA750040



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, FC
Molecular Wt:	Predicted band size: 41 kDa
Clone number:	SR43-04

Description: p38 mitogen-activated protein kinases are a class of mitogen-activated protein kinases (MAPKs) that are responsive to stress stimuli, such as cytokines, ultraviolet irradiation, heat shock, and osmotic shock, and are involved in cell differentiation, apoptosis and autophagy. Persistent activation of the p38 MAPK pathway in muscle satellite cells (muscle stem cells) due to ageing, impairs muscle regeneration. p38 MAP Kinase (MAPK), also called RK or CSBP (Cytokinin Specific Binding Protein), is the mammalian orthologue of the yeast Hog1p MAP kinase, which participates in a signaling cascade controlling cellular responses to cytokines and stress. Four p38 MAP kinases, p38- α (MAPK14), - β (MAPK11), - γ (MAPK12 / ERK6), and - δ (MAPK13 / SAPK4), have been identified. Similar to the SAPK/JNK pathway, p38 MAP kinase is activated by a variety of cellular stresses including osmotic shock, inflammatory cytokines, lipopolysaccharides (LPS), ultraviolet light, and growth factors. MKK3 and SEK activate p38 MAP kinase by phosphorylation at Thr-180 and Tyr-182.

Immunogen: Synthetic peptide within human p38 aa 160-200.

Positive control: Hela cell lysate, NIH/3T3 cell lysate, PC-12 cell lysate, THP-1 cell lysate, Jurkat cell lysate, C2C12 cell lysate, rat kidney tissue lysate, mouse kidney tissue lysate, HeLa, RAW264.7, C6.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: Q16539 Human | P47811 Mouse | P70618 Rat

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:100-1:500
FC	1:1,000

Storage Buffer: 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.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

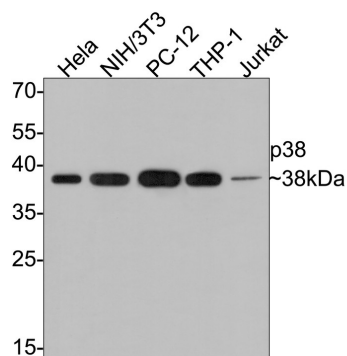


Fig1: Western blot analysis of p38 alpha / MAPK14 on different lysates with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/1,000 dilution.

Lane 1: HeLa cell lysate
 Lane 2: NIH/3T3 cell lysate
 Lane 3: PC-12 cell lysate
 Lane 4: THP-1 cell lysate
 Lane 5: Jurkat cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 41 kDa
 Observed band size: 38 kDa

Exposure time: 2 minutes;
 12% 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 (HA750040) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

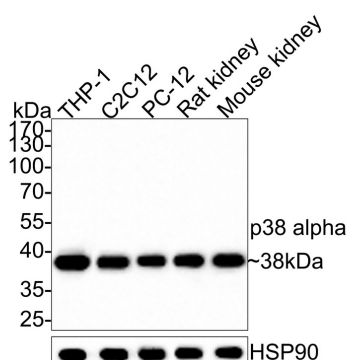


Fig2: Western blot analysis of p38 alpha / MAPK14 on different lysates with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/1,000 dilution.

Lane 1: THP-1 cell lysate (10 µg/Lane)
 Lane 2: C2C12 cell lysate (10 µg/Lane)
 Lane 3: PC-12 cell lysate (10 µg/Lane)
 Lane 4: Rat kidney tissue lysate (20 µg/Lane)
 Lane 5: Mouse kidney tissue lysate (20 µg/Lane)

Predicted band size: 41 kDa
 Observed band size: 38 kDa

Exposure time: 3 minutes 10 seconds;
 10% 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 (HA750040) at 1/1,000 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.

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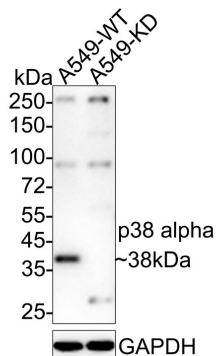
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Fig3: Western blot analysis of p38 alpha / MAPK14 on different lysates with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/2,000 dilution.

Lane 1: A549-WT cell lysate

Lane 2: A549-KD p38 alpha / MAPK14 cell lysate



Lysates/proteins at 10 µg/Lane.

Predicted band size: 41 kDa

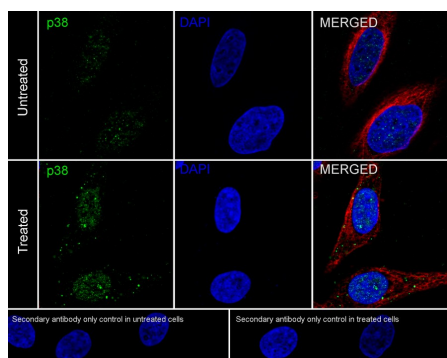
Observed band size: 38 kDa

Exposure time: 3 minutes; ECL: K1801;

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 (HA750040) 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.

Fig4: Immunocytochemistry analysis of HeLa cells treated with UV for 30 minutes then recover 30 minutes labeling p38 alpha / MAPK14 with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/500 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/500 dilution in 1% BSA in PBST overnight at 4°C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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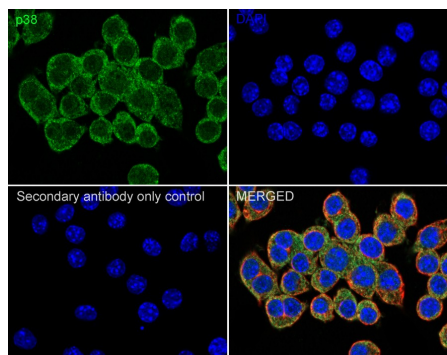
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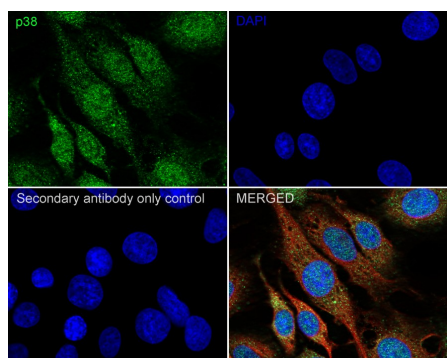
Fig5: Immunocytochemistry analysis of RAW264.7 cells labeling p38 alpha / MAPK14 with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Fig6: Immunocytochemistry analysis of C6 cells labeling p38 alpha / MAPK14 with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/100 dilution.



Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-p38 alpha / MAPK14 antibody (HA750040) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

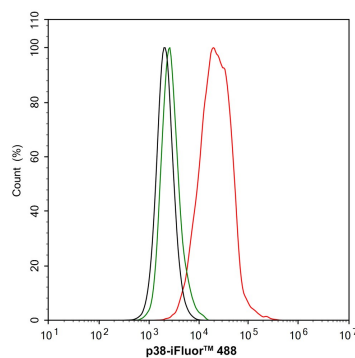


Fig7: Flow cytometric analysis of HeLa cells labeling p38 alpha / MAPK14.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750040, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4°C for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4°C. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

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

Background References

1. Yan T et al. Luteolin inhibits behavioral sensitization by blocking methamphetamine-induced MAPK pathway activation in the caudate putamen in mice. PLoS One 9:e98981 (2014).
2. Pagliara V et al. Protease Nexin-1 affects the migration and invasion of C6 glioma cells through the regulation of urokinase Plasminogen Activator and Matrix Metalloproteinase-9/2. Biochim Biophys Acta 1843:2631-44 (2014).

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