

Anti-SIRP alpha Antibody [JE36-20]

HA721316



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, FC
Molecular Wt:	Predicted band size: 55 kDa
Clone number:	JE36-20

Description: Immunoglobulin-like cell surface receptor for CD47. Acts as docking protein and induces translocation of PTPN6, PTPN11 and other binding partners from the cytosol to the plasma membrane. Supports adhesion of cerebellar neurons, neurite outgrowth and glial cell attachment. May play a key role in intracellular signaling during synaptogenesis and in synaptic function. Involved in the negative regulation of receptor tyrosine kinase-coupled cellular responses induced by cell adhesion, growth factors or insulin. Mediates negative regulation of phagocytosis, mast cell activation and dendritic cell activation. CD47 binding prevents maturation of immature dendritic cells and inhibits cytokine production by mature dendritic cells. Ubiquitous. Highly expressed in brain. Detected on myeloid cells, but not T-cells. Detected at lower levels in heart, placenta, lung, testis, ovary, colon, liver, small intestine, prostate, spleen, kidney, skeletal muscle and pancreas. Binds PTPN11 when tyrosine-phosphorylated, except in macrophages, where it primarily binds PTPN6. Binds GRB2 in vitro. Binds FGR. Binds JAK2 irrespective of its phosphorylation status and forms a stable complex. Binds SCAP1 and/or SCAP2. The resulting complex recruits FYB1. Binds PTK2B.

Immunogen: Recombinant protein within Human SIRP alpha aa 395-504 (Cytoplasmic).

Positive control: THP-1 cell lysate, HL-60 cell lysate, mouse brain tissue lysate, human lymph node tissue, human spleen tissue.

Subcellular location: Membrane

Database links: SwissProt: P78324 Human | P97797 Mouse | P97710 Rat

Recommended Dilutions:

WB	1:1,000
IHC-P	1:1,000
FC	1:1,000

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein A affinity purified.

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Orders:0086-571-88062880

Technical:0086-571-89986345

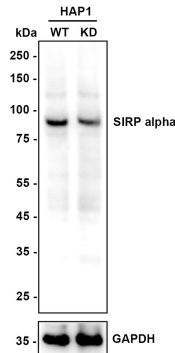
Service mail:support@huabio.cn

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Images

Fig1: Western blot analysis of SIRP alpha on different lysates with Rabbit anti-SIRP alpha antibody (HA721316) at 1/1,000 dilution.

Lane 1: HAP1-parental cell lysate
Lane 2: HAP1-SIRP alpha KD cell lysate



Lysates/proteins at 10 µg/Lane.

Predicted band size: 55 kDa
Observed band size: 75-100 kDa

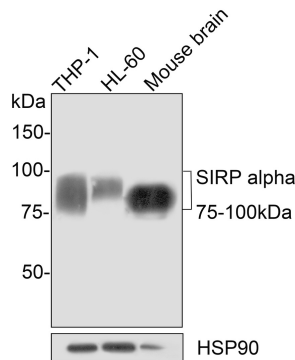
Exposure time: 9 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDN/TBST for 1 hour at room temperature. The primary antibody (HA721316) at 1/1,000 dilution was used in K1803 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: Western blot analysis of SIRP alpha on different lysates with Rabbit anti-SIRP alpha antibody (HA721316) at 1/1,000 dilution.

Lane 1: THP-1 cell lysate (10 µg/Lane)
Lane 2: HL-60 cell lysate (10 µg/Lane)
Lane 3: Mouse brain tissue lysate (20 µg/Lane)



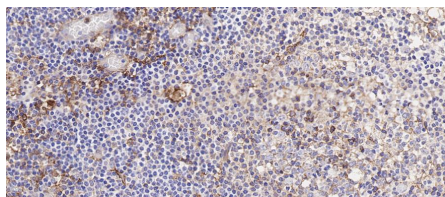
Predicted band size: 55 kDa
Observed band size: 75-100 kDa

Exposure time: 2 minutes;

8% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDN/TBST for 1 hour at room temperature. The primary antibody (HA721316) at 1/1,000 dilution was used in 5% NFDN/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.

Fig3: Immunohistochemical analysis of paraffin-embedded human lymph node tissue with Rabbit anti-SIRP alpha antibody (HA721316) at 1/1,000 dilution.



The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were

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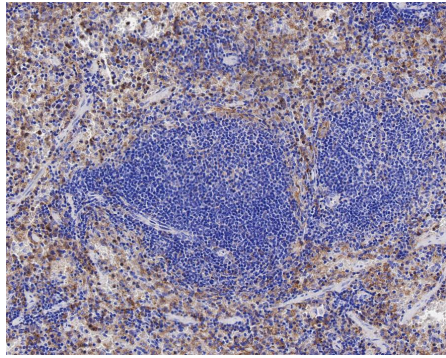


Fig4: Immunohistochemical analysis of paraffin-embedded human spleen tissue with Rabbit anti-SIRP alpha antibody (HA721316) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 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 (HA721316) at 1/1,000 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.

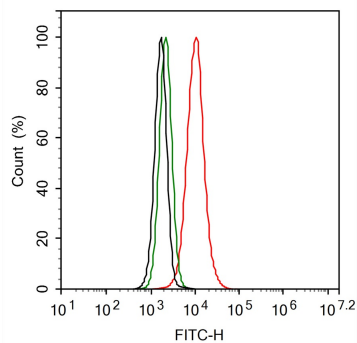


Fig5: Flow cytometric analysis of THP-1 cells labeling SIRP alpha.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA721316, 1ug/ml) (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 Alexa Fluor® 488 conjugate-Goat anti-Rabbit IgG Secondary antibody 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. Timms J.F., Swanson K.D., Marie-Cardine A., Raab M., Rudd C.E., Schraven B., Neel B.G. SHPS-1 is a scaffold for assembling distinct adhesion-regulated multi- protein complexes in macrophages. *Curr. Biol.* 9:927-930 (1999).
2. Latour S., Tanaka H., Demeure C., Mateo V., Rubio M., Brown E.J., Maliszewski C., Lindberg F.P., Oldenborg A., Sarfati M. Bidirectional negative regulation of human T and dendritic cells by CD47 and its cognate receptor signal-regulator protein-alpha: down-regulation of IL-12 responsiveness and inhibition of dendritic cell activation. *J. Immunol.* 167:2547-2554 (2001)

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