

Anti-STAT6 Antibody [SY02-72]

ET1607-61



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP
Molecular Wt:	Predicted band size: 94 kDa
Clone number:	SY02-72

Description: Signal transducer and activator of transcription 6 (STAT6) is a transcription factor that belongs to the Signal Transducer and Activator of Transcription (STAT) family of proteins. The proteins of STAT family transmit signals from a receptor complex to the nucleus and activate gene expression. Similarly as other STAT family proteins, STAT6 is also activated by growth factors and cytokines. STAT6 is mainly activated by cytokines interleukin-4 and interleukin-13. STAT6-mediated signaling pathway is required for the development of T-helper type 2 (Th2) cells and Th2 immune response. Activation of STAT6 signaling pathway is necessary in macrophage function, and is required for the M2 subtype activation of macrophages. STAT6 is also involved in IL4 signaling in B cells, and STAT6 determines the levels of CD20 on the surface of normal and malignant B lymphocytes. STAT6 also plays a critical role in Th2 lung inflammatory responses including clearance of parasitic infections and in the pathogenesis of asthma. Th2-cell derived cytokines as IL-4 and IL-13 induce the production of IgE which is a major mediator in allergic response.

Immunogen: Synthetic peptide within human STAT6 C terminal.

Positive control: HeLa cell lysate, Raji cell lysate, HeLa, AGS, NIH/3T3, human breast carcinoma tissue, human kidney tissue, mouse lung tissue, human tonsil tissue, mouse stomach tissue.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: P42226 Human | P52633 Mouse

Recommended Dilutions:

WB	1:500
IF-Cell	1:50
IF-Tissue	1:50
IHC-P	1:50-1:200
IP	Use at an assay dependent concentration.

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 or -80°C. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

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Images

Fig1: Western blot analysis of STAT6 on different lysates with Rabbit anti-STAT6 antibody (ET1607-61) at 1/500 dilution.

Lane 1: HeLa cell lysate

Lane 2: Raji cell lysate

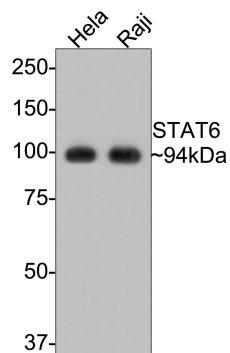
Lysates/proteins at 10 µg/Lane.

Predicted band size: 94 kDa

Observed band size: 94 kDa

Exposure time: 30 seconds;

8% 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 (ET1607-61) 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 1:300,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of STAT6 on different lysates with Rabbit anti-STAT6 antibody (ET1607-61) at 1/500 dilution.

Lane 1: HeLa-si-NT cell lysate

Lane 2: HeLa-si-STAT6 cell lysate

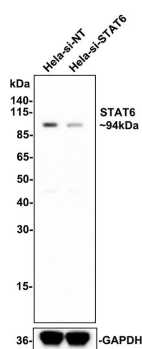
Lysates/proteins at 10 µg/Lane.

Predicted band size: 94 kDa

Observed band size: 94 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.



ET1607-61 was shown to specifically react with STAT6 in HeLa-si-NT cells. Weakened band was observed when HeLa-si-STAT6 sample was tested. HeLa-si-NT and HeLa-si-STAT6 samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDm in TBST for 1 hour at room temperature. The primary antibody (ET1607-61, 1/500) and Loading control antibody (Rabbit anti-GAPDH, ET1601-4, 1/10,000) were used in 5% BSA 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.

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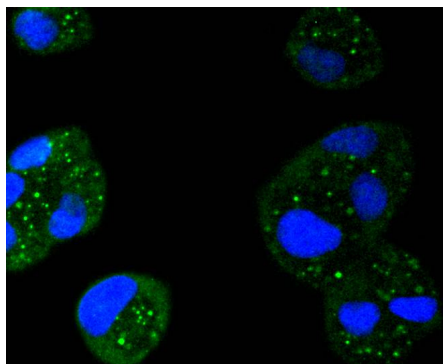


Fig3: Immunocytochemistry analysis of HeLa cells labeling STAT6 with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 °C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution in 2% negative goat serum 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.

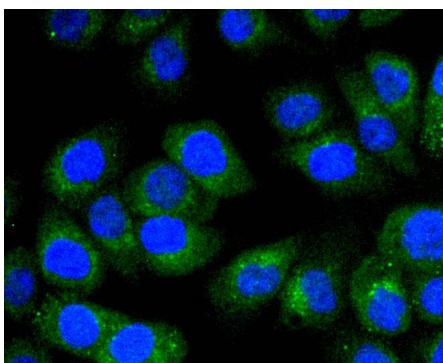


Fig4: Immunocytochemistry analysis of AGS cells labeling STAT6 with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 °C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution in 2% negative goat serum 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.

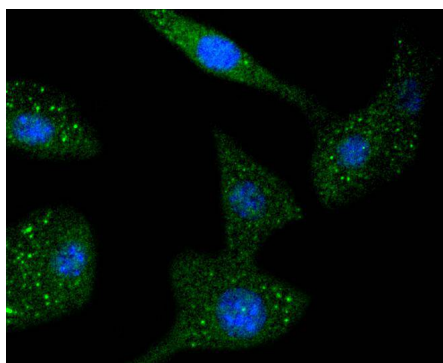


Fig5: Immunocytochemistry analysis of NIH/3T3 cells labeling STAT6 with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 °C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution in 2% negative goat serum 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.

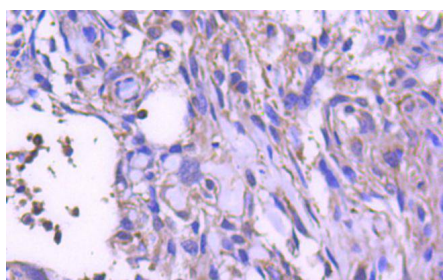


Fig6: Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution.

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 (ET1607-61) at 1/50 dilution in 2% negative goat serum 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.

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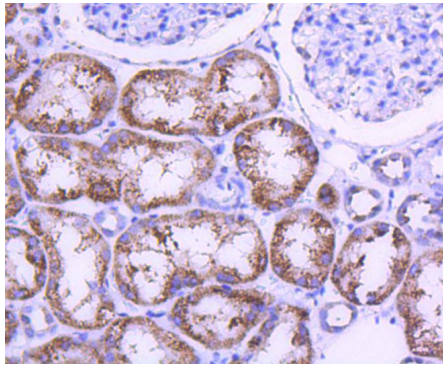


Fig7: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution.

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 (ET1607-61) at 1/50 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.

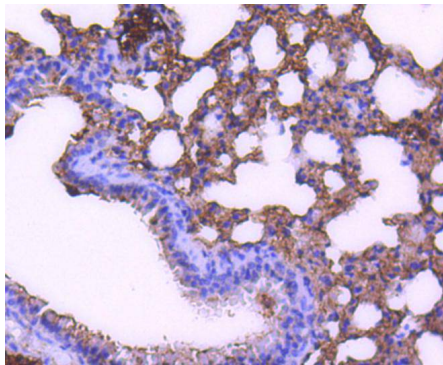


Fig8: Immunohistochemical analysis of paraffin-embedded mouse lung tissue with Rabbit anti-STAT6 antibody (ET1607-61) at 1/20 dilution.

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 (ET1607-61) at 1/20 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.

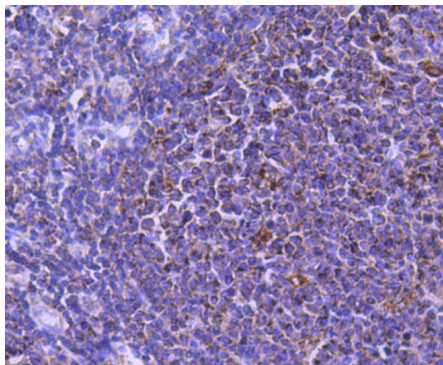


Fig9: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution.

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 (ET1607-61) at 1/50 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.

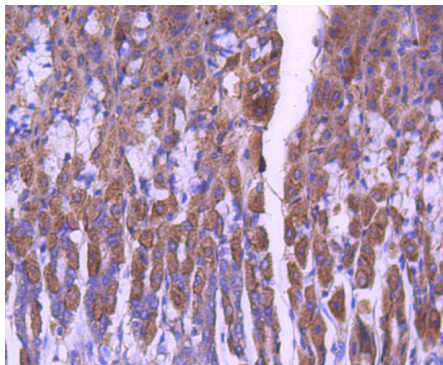


Fig10: Immunohistochemical analysis of paraffin-embedded mouse stomach tissue with Rabbit anti-STAT6 antibody (ET1607-61) at 1/50 dilution.

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 (ET1607-61) at 1/50 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. Zheng, C. et al. 2015. CD11b regulates obesity-induced insulin resistance via limiting alternative activation and proliferation of adipose tissue macrophages. *Proc. Natl. Acad. Sci. U.S.A.*. 112: E7239-48.
2. Carlson, T.J. et al. 2014. Halofuginone-induced amino acid starvation regulates Stat3-dependent Th17 effector function and reduces established autoimmune inflammation. *J. Immunol.*. 192: 2167-76.

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