## Anti-STAT3 Antibody [SY24-08] - BSA and Azide free HA750120

Product Type: Recombinant Rabbit monoclonal IgG, primary antibodies

Species reactivity: Human, Mouse, Rat, Zebrafish

Applications: WB, IF-Cell, IHC-P, FC

Molecular Wt: Predicted band size: 88 kDa

Clone number: SY24-08

**Description:** Signal transducer and transcription activator that mediates cellular responses to interleukins,

KITLG/SCF, LEP and other growth factors. Once activated, recruits coactivators, such as NCOA1 or MED1, to the promoter region of the target gene. May mediate cellular responses to activated FGFR1, FGFR2, FGFR3 and FGFR4. Upon activation of IL6ST/gp130 signaling by interleukin-6 (IL6), binds to the IL6-responsive elements identified in the promoters of various acute-phase protein genes. Activated by IL31 through IL31RA. Acts as a regulator of inflammatory response by regulating differentiation of naive CD4(+) T-cells into T-helper Th17 or regulatory T-cells (Treg): deacetylation and oxidation of lysine residues by LOXL3, leads to disrupt STAT3 dimerization and inhibit its transcription activity. Involved in cell cycle regulation by inducing the expression of key genes for the progression from G1 to S phase, such as CCND1. Mediates the effects of LEP on melanocortin production, body energy homeostasis and lactation (By similarity). May play an apoptotic role by transctivating BIRC5 expression under LEP activation. Cytoplasmic STAT3 represses macroautophagy by inhibiting EIF2AK2/PKR activity. Plays a crucial role in basal beta cell functions, such as

regulation of insulin secretion. Shuttles between the nucleus and the cytoplasm.

**Immunogen:** Synthetic peptide within human Stat3 aa 670-710.

Positive control: CRC, HeLa cell lysate, A431 cell lysate, PANC-1 cell lysate, NIH/3T3 cell lysate, RAW264.7

cell lysate, PC-12 cell lysate, C6 cell lysate, mouse brain tissue, mouse pancreas tissue, rat

brain tissue, rat pancreas tissue.

Subcellular location: Cytoplasm, Nucleus.

Database links: SwissProt: P40763 Human | P42227 Mouse | P52631 Rat

**Recommended Dilutions:** 

WB 1:2,000 IF-Cell 1:50-1:200 IHC-P 1:50-1:200 FC 1μg/mL

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.

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## **Images**

Fig1: Western blot analysis of STAT3 on different lysates with Rabbit anti-STAT3 antibody (HA750120) at 1/2,000 dilution.

Lane 1: Hela-si NT cell lysate Lane 2: Hela-si STAT3 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 88 kDa Observed band size: 88 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ET1607-38, 1/2,000) 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 1/100,000 dilution was used for 1 hour at room temperature.

**Fig2:** Western blot analysis of STAT3 on different lysates with Rabbit anti-STAT3 antibody (HA750120) at 1/2,000 dilution.

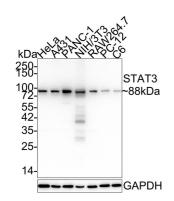
Lane 1: HeLa cell lysate Lane 2: A431 cell lysate Lane 3: PANC-1 cell lysate Lane 4: NIH/3T3 cell lysate Lane 5: RAW264.7 cell lysate Lane 6: PC-12 cell lysate Lane 7: C6 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 88 kDa Observed band size: 88 kDa

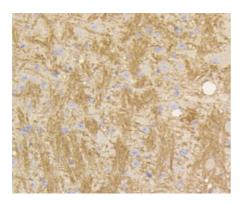
Exposure time: 2 minutes 24 seconds;

4-20% SDS-PAGE gel.

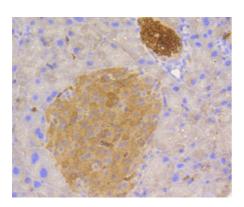


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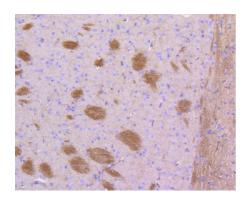




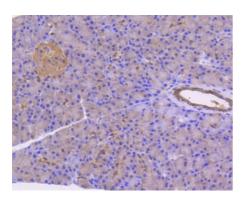
**Fig3:** Immunohistochemical analysis of paraffin-embedded mouse brain tissue using anti-STAT3 antibody. The section was pretreated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA750120, 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.



**Fig4:** Immunohistochemical analysis of paraffin-embedded mouse pancreas tissue using anti-STAT3 antibody. The section was pretreated 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 ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA750120, 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.



**Fig5**: Immunohistochemical analysis of paraffin-embedded rat brain tissue using anti-STAT3 antibody. The section was pretreated 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 ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA750120, 1/200) 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.



**Fig6:** Immunohistochemical analysis of paraffin-embedded rat pancreas tissue using anti-STAT3 antibody. The section was pretreated 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 ddH $_2$ O and PBS, and then probed with the primary antibody (HA750120, 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.

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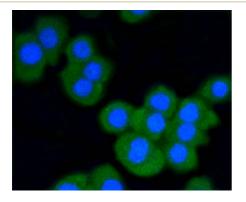


Fig7: ICC staining of STAT3 in CRC cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750120, 1/50) for 1 hour at room 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).

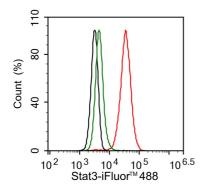


Fig8: Flow cytometric analysis of HeLa cells labeling STAT3.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA750120,  $1\mu g/mL$ ) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at  $+4^{\circ}C$  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  $+4^{\circ}C$ . 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. Tsugawa D et al. Specific activin receptor-like kinase 3 inhibitors enhance liver regeneration. J Pharmacol Exp Ther 351:549-58 (2014).
- 2. Wu Q et al. Crosstalk of JNK1-STAT3 is critical for RAW264.7 cell survival. Cell Signal 26:2951-60 (2014).