Phospho-STAT3-S727 Rabbit mAb

Catalog No.: AP1593 Recombinant



Basic Information

Observed MW

91 kDa

Calculated MW

88 kDa/83 kDa

Category

Primary antibody

Applications

WB,IP,IF/ICC,IHC-P,ChIP,ELISA

Cross-Reactivity

Human, Mouse, Rat

CloneNo number

ARC75347

Background

The protein encoded by this gene is a member of the STAT protein family. In response to cytokines and growth factors, STAT family members are phosphorylated by the receptor associated kinases, and then form homo- or heterodimers that translocate to the cell nucleus where they act as transcription activators. This protein is activated through phosphorylation in response to various cytokines and growth factors including IFNs, EGF, IL5, IL6, HGF, LIF and BMP2. This protein mediates the expression of a variety of genes in response to cell stimuli, and thus plays a key role in many cellular processes such as cell growth and apoptosis. The small GTPase Rac1 has been shown to bind and regulate the activity of this protein. PIAS3 protein is a specific inhibitor of this protein. This gene also plays a role in regulating host response to viral and bacterial infections. Mutations in this gene are associated with infantile-onset multisystem autoimmune disease and hyper-immunoglobulin E syndrome.

Recommended Dilutions

WB 1:8000 - 1:16000

0.5μg-4μg antibody for ΙP

200µg-400µg extracts of whole cells

IF/ICC 1:1000 - 1:5000

IHC-P 1:3000 - 1:12000

ChIP 3µg antibody for 10μg-15μg of Chromatin

Recommended starting

ELISA

concentration is 1 µg/mL. Please optimize the concentration based on your specific assay requirements. For highratio antibody dilutions (≥1:10000)∏a sequential dilution method is strongly recommended to ensure measurement

accuracy.

Immunogen Information

Gene ID Swiss Prot 6774 P40763

Immunogen

Synthetic peptide. This information is considered to be commercially sensitive.

Synonyms

APRF; HIES; ADMIO; ADMIO1

Product Information

Source Isotype **Purification** Rabbit IgG Affinity purification

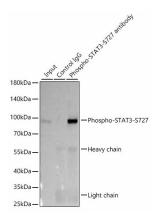
Storage

Store at -20°C. Avoid freeze / thaw cycles.

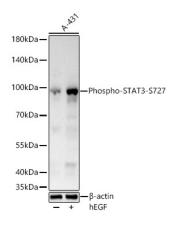
Buffer: PBS with 0.09% Sodium azide, 0.05% BSA, 50% glycerol, pH7.3.

Contact

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•	www.abclonal.com.cr



Immunoprecipitation of Phospho-STAT3-S727 from 300 μg extracts of HeLa cells was performed using 1 μg of Phospho-STAT3-S727 Rabbit mAb (AP1593). Rabbit Control IgG (AC005) was used to precipitate the Control IgG sample. IP samples were eluted with 1x reducing Laemmli Buffer. The Input lane represents 10% of the total input. Western blot analysis of immunoprecipitates was conducted using Phospho-STAT3-S727 Rabbit mAb (AP1593) at a dilution of 1:20000.



Western blot analysis of lysates from A-431 cells using Phospho-STAT3-S727 Rabbit mAb (AP1593) at 1:16000 dilution incubated overnight at 4° C. A-431 cells were treated with hEGF (100 ng/mL) at 37° C for 30 minutes after serum-starvation overnight.

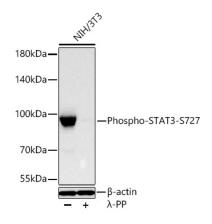
Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.

Lysates/proteins: 30 μg per lane.

Blocking buffer: 3% nonfat dry milk in TBST.

Detection: ECL Basic Kit (RM00020).

Exposure time: 90 s.



Western blot analysis of lysates from NIH/3T3 cells using Phospho-STAT3-S727 Rabbit mAb (AP1593) at 1:16000 dilution incubated overnight at 4°C. NIH/3T3 cells were treated with λ -PP mixed solution (2 U/ μ L) at 37°C for 30 minutes.

Secondary antibody: HRP-conjugated Goat anti-Rabbit IgG (H+L) (AS014) at 1:10000 dilution.

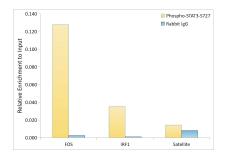
Lysates/proteins: 30 µg per lane.

Blocking buffer: 3% nonfat dry milk in TBST.

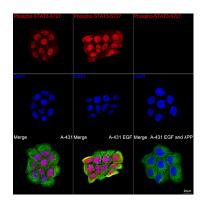
Detection: ECL Basic Kit (RM00020).

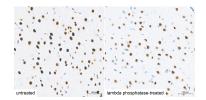
Exposure time: 60 s.

Validation Data



Chromatin immunoprecipitation was performed with 15 μ g of cross-linked chromatin from HeLa cells starved overnight and treaded with IFN- α (50ng/mL,15mins), using 3 μ g of Phospho-STAT3-S727 Rabbit mAb (AP1593) and Rabbit IgG isotype control (AC042). The enrichment of immunoprecipitated DNA at different genomic loci was examined by quantitative PCR. The histogram compares the ratio of the immunoprecipitated DNA to the input at given loci.





Confocal imaging of A-431 cells(untreated), A-431 cells(treated with EGF) and A-431 cells(treated with EGF) and A-431 cells(treated with EGF and $\lambda pp)$ using Phospho-STAT3-S727 Rabbit mAb (AP1593, dilution 1:5000) followed by a further incubation with Cy3-conjugated Goat Anti-Rabbit 1gG (H+L) (AS007, dilution 1:500) (Red). The cells were counterstained with α -Tubulin Mouse mAb (AC012, dilution 1:400) followed by incubation with ABflo® 488-conjugated Goat Anti-Mouse 1gG (H+L) (AS076, dilution 1:500) (Green). DAPI was used for nuclear staining (Blue). Objective: 100x.

Immunohistochemistry analysis of paraffinembedded Rat brain tissue, untreated (left) and lambda phosphatase-treated (right), using Phospho-STAT3-S727 Rabbit mAb (AP1593) at a dilution of 1:9000 (40x lens). High pressure antigen retrieval performed with 0.01M Tris-EDTA Buffer (pH 9.0) prior to IHC staining.