# abcam

### **Product datasheet**

## Anti-STAT1 (phospho S727) antibody [EPR3146] ab109461

重组 RabMAb

性能

★★★★★ <u>1 Abreviews</u> <u>38 References</u> 11 图像

概述	
产品名称	Anti-STAT1 (phospho S727) <b>抗体</b> [EPR3146]
描述	免单克隆抗体[EPR3146] to STAT1 (phospho S727)
宿主	Rabbit
特异性	A phospho specific peptide corresponding to residues surrounding Serine 727 of human Stat-1 was used as an immunogen. This antibody only detects Stat-1 phosphorylated at Serine 727.
经测试应 <b>用</b>	适用于: WB, IHC-P, ChIC/CUT&RUN-seq, Dot blot 不适用于: ICC/IF
<b>种属反</b> 应性	<b>与反</b> 应: Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
<b>阳性</b> 对 <b>照</b>	WB: HeLa cell lysate. Rat and mouse brain lysate. IHC-P: Rat and mouse colon tissue. Human breast carcinoma and stomach adenocarcinoma tissue. ChIC/CUT&RUN-Seq: HeLa cells.
<b>常</b> 规说 <b>明</b>	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information <u>see here</u> . Our RabMAb <sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <u>RabMAb<sup>®</sup> patents</u> .

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.
	Avoid freeze / thaw cycle.
存储溶液	pH: 7.20
	Preservative: 0.01% Sodium azide Constituents: 40% Glycerol, 59% PBS, 0.05% BSA
纯 <b>度</b>	Protein A purified
克隆	单 <b>克隆</b>

同种型

lgG

#### 应用

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#### "应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	<b>★★★★</b>	1/1000 - 1/10000. Detects a band of approximately 91 kDa (predicted molecular weight: 87 kDa).
IHC-P		1/100 - 1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Heat up to 98 °C, below boiling, and then let cool for 10-20 minutes.
ChIC/CUT&RUN-seq		Use at an assay dependent concentration. 5µg
Dot blot		Use at an assay dependent concentration.

#### 应用说明

Is unsuitable for ICC/IF.

#### 靶标

功能

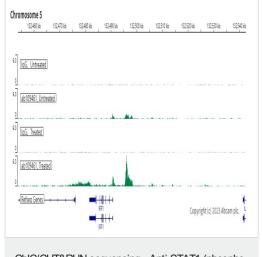
Signal transducer and activator of transcription that mediates signaling by interferons (IFNs). Following type I IFN (IFN-alpha and IFN-beta) binding to cell surface receptors, Jak kinases (TYK2 and JAK1) are activated, leading to tyrosine phosphorylation of STAT1 and STAT2. The phosphorylated STATs dimerize, associate with ISGF3G/IRF-9 to form a complex termed ISGF3 transcription factor, that enters the nucleus. ISGF3 binds to the IFN stimulated response element (ISRE) to activate the transcription of interferon stimulated genes, which drive the cell in an antiviral state. In response to type II IFN (IFN-gamma), STAT1 is tyrosine- and serinephosphorylated. It then forms a homodimer termed IFN-gamma-activated factor (GAF), migrates into the nucleus and binds to the IFN gamma activated sequence (GAS) to drive the expression of the target genes, inducing a cellular antiviral state.

疾病相关

Note=STAT1 deficiency results in impaired immune response leading to severe mycobacterial and viral diseases. In the case of complete deficiency, patients can die of viral disease. Defects in STAT1 are a cause of mendelian susceptibility to mycobacterial disease (MSMD) [MIM:209950]; also known as familial disseminated atypical mycobacterial infection. This rare condition confers predisposition to illness caused by moderately virulent mycobacterial species, such as Bacillus Calmette-Guerin (BCG) vaccine and environmental non-tuberculous mycobacteria, and by the more virulent Mycobacterium tuberculosis. Other microorganisms rarely cause severe clinical disease in individuals with susceptibility to mycobacterial infections, with the exception of Salmonella which infects less than 50% of these individuals. The pathogenic mechanism underlying MSMD is the impairment of interferon-gamma mediated immunity whose severity determines the clinical outcome. Some patients die of overwhelming mycobacterial disease with lepromatous-like lesions in early childhood, whereas others develop, later in life,

	disseminated but curable infections with tuberculoid granulomas. MSMD is a genetically heterogeneous disease with autosomal recessive, autosomal dominant or X-linked inheritance.
序列相似性	Belongs to the transcription factor STAT family. Contains 1 SH2 domain.
<b>翻</b> 译后 <b>修</b> 饰	<ul> <li>Phosphorylated on tyrosine and serine residues in response to IFN-alpha, IFN-gamma, PDGF and EGF. Phosphorylation on Tyr-701 (lacking in beta form) by JAK promotes dimerization and subsequent translocation to the nucleus. Phosphorylation on Ser-727 by several kinases including MAPK14, ERK1/2 and CAMKII on IFN-gamma stimulation, regulates STAT1 transcriptional activity. Phosphorylation on Ser-727 promotes sumoylation though increasing interaction with PIAS. Phosphorylation on Ser-727 by PKCdelta induces apoptosis in response to DNA-damaging agents.</li> <li>Sumoylated by SUMO1, SUMO2 and SUMO3. Sumoylation is enhanced by IFN-gamma-induced phosphorylation on Ser-727, and by interaction with PIAS proteins. Enhances the transactivation activity.</li> <li>ISGylated.</li> </ul>
细 <b>胞定位</b>	Cytoplasm. Nucleus. Translocated into the nucleus in response to IFN-gamma-induced tyrosine phosphorylation and dimerization.

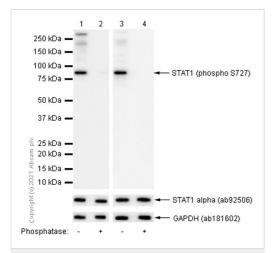
#### 图片



ChIC/CUT&RUN sequencing - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) ChIC/CUT&RUN was performed using a pAG-MNAse at a final concentration of 700 ng/mL, 2 x 10^5 HeLa (Human cervix adenocarcinoma epithelial cell line) treated with IFN gamma (50ng/ml 1h) cells and 5µg of ab109461 [EPR3146]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control **ab172730** is also shown.

Additional screenshots of mapped reads can be downloaded here.

The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



Western blot - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) All lanes : Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) at 1/1000 dilution

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate (treated with Alkaline Phosphatase for 1 hour)

Lane 3 : Mouse brain lysate

Lane 4 : Mouse brain lysate (treated with Alkaline Phosphatase for 1 hour)

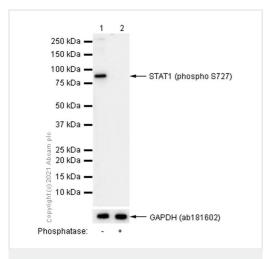
Lysates/proteins at 15 µg per lane.

#### Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 87 kDa Observed band size: 91 kDa

Blocking buffer: 5% NFDM/TBST.



Western blot - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) All lanes : Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) at 1/1000 dilution

Lane 1 : Rat brain lysate Lane 2 : Rat brain lysate (treated with Alkaline Phosphatase for 1 hour)

Lysates/proteins at 15 µg per lane.

### Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 87 kDa Observed band size: 91 kDa

#### Blocking buffer: 5% NFDM/TBST.

**All lanes :** Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) at 1/5000 dilution (purified)

Lane 1 : Untreated HeLa whole cell lysate

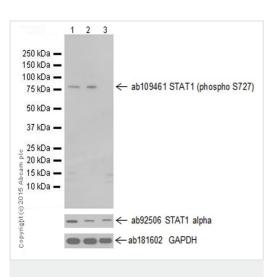
Lane 2 : HeLa whole cell lysate treated with etoposide

Lane 3 : HeLa whole cell lysate treated with etoposide, followed by membrane treatment with phosphatase

Lysates/proteins at 10 µg per lane.

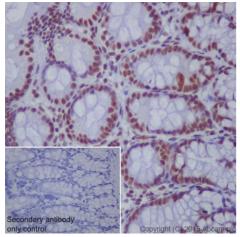
#### Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution



Western blot - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) Predicted band size: 87 kDa Observed band size: 90 kDa

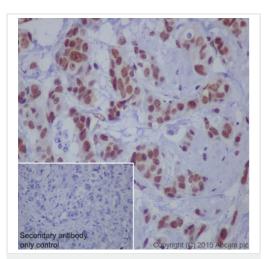
Blocking buffer: 2% BSA/TBST Dilution buffer: 2% BSA/TBST



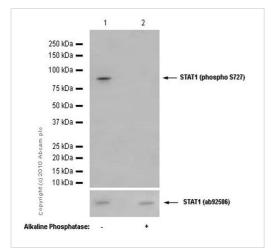
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461)

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) Immunohistochemical staining of paraffin embedded rat colon with purified ab109461 at a working dilution of 1/200. The secondary antibody used is <u>ab97051</u>, a goat anti-rabbit IgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.

Immunohistochemical staining of paraffin embedded mouse colon with purified ab109461 at a working dilution of 1/200. The secondary antibody used is **ab97051**, a goat anti-rabbit IgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) Immunohistochemical staining of paraffin embedded human breast carcinoma with purified ab109461 at a working dilution of 1/200. The secondary antibody used is <u>ab97051</u>, a goat anti-rabbit IgG (H&L) at a dilution of 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Western blot - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) **All lanes :** Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) at 1/10000 dilution (unpurified)

Lane 1 : Untreated HeLa (human cervix adenocarcinoma) Lane 2 : HeLa (human cervix adenocarcinoma) membrane treated with Alkaline Phosphatase

Lysates/proteins at 10 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1500 dilution

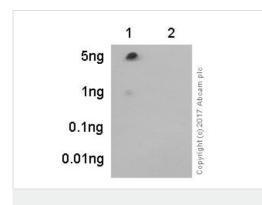
Developed using the ECL technique.

Predicted band size: 87 kDa Observed band size: 91 kDa

Exposure time: 30 seconds

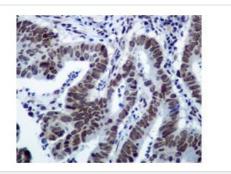
The lower section shows STAT1 detected with <u>ab92506</u>, anti-STAT1 antibody, to confirm that the same amount of lysate is used in each lane.

Blocking and dilution biffer: 5% NFDM/TBST.



Dot Blot - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461)

Dot Blot analysis of Lane 1: STAT1 (pS727) phospho peptide and Lane 2: STAT1 non-phospho peptide, labeling STAT1 (phospho S727) with ab109461 at 1/1000 dilution. 5% NFDM/TBST was used as the blocking and diluting buffer. **ab97051**, a Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated secondary antibody was used at 1/100000 dilution. Exposure time: 3 minutes.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461)



Anti-STAT1 (phospho S727) antibody [EPR3146] (ab109461) Unpurified ab109461, at a 1/100 dilution, staining STAT1 (phospho S727) in paraffin embedded Human stomach adenocarcinoma tissue by Immunohistochemistry.

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