abcam

Product datasheet

Human STAT3 knockout HeLa cell lysate ab263797

画像数 4

製品の概要

製品名	Human STAT3 knockout HeLa cell lysate
製品の概要	Knockout cell lysate achieved by CRISPR/Cas9.
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 1 bp deletion in exon2.
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Reconstitution notes	To use as WB control, resuspend the lyophilizate in 50 µL of LDS* Sample Buffer to have a final concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M DTT. **Usage of SDS sample buffer is not recommended with these lyophilized lysates.*
特記事項	**Lysate preparation:** Our lysates are made using RIPA buffer to which we add a protease inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found **here**. Please refer to our lysis protocol for further details on how our lysates are prepared. **User storage instructions:** Lyophilizate may be stored at 4°C. After reconstitution, store at -20°C for short-term storage or -80°C for long-term storage. Access thousands of knockout cell lysates, generated from commonly used cancer cell lines. **See here for more information on knockout cell lysates.** Abcam has not and does not intend to apply for the REACH Authorisation of customers' uses of products that contain European Authorisation list (Annex XIV) substances. It is the responsibility of our customers to check the necessity of application of REACH Authorisation, and any other relevant authorisations, for their intended uses. This product is subject to limited use licenses from The Broad Institute and ERS Genomics Limited, and is developed with patented technology. For full details of the limited use licenses and relevant patents please refer to our **limited use license** and **patent pages**.
アプリケーション	**適用あり:** WB

製品の特性

保存方法 Store at -80°C. Please refer to protocols.

内容	1 kit
ab255540 - Human STAT3 knockout HeLa cell lysate	1 x 100µg
ab255552 - Human wild-type HeLa cell lysate	1 x 100µg

Cell type epithelial

Disease Adenocarcinoma

Gender Female

STR Analysis Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8, 12 CSF1PO: 9, 10

ターゲット情報

機能 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 (PubMed:17344214). May mediate cellular responses to activated FGFR1, FGFR2, FGFR3 and FGFR4. Binds to the interleukin-6 (IL-6)-responsive elements identified in the promoters of various acute-phase protein genes. Activated by IL31 through IL31RA. Involved in cell cycle regulation by inducing the expression of key genes for the progression from G1 to S phase, such as CCND1 (PubMed:17344214). 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 (PubMed:18242580). Cytoplasmic STAT3 represses macroautophagy by inhibiting EIF2AK2/PKR activity.

組織特異性 Heart, brain, placenta, lung, liver, skeletal muscle, kidney and pancreas.

関連疾患 Hyperimmunoglobulin E recurrent infection syndrome, autosomal dominant
Autoimmune disease, multisystem, infantile-onset

配列類似性 Belongs to the transcription factor STAT family.
Contains 1 SH2 domain.

翻訳後修飾 Tyrosine phosphorylated upon stimulation with EGF. Tyrosine phosphorylated in response to constitutively activated FGFR1, FGFR2, FGFR3 and FGFR4 (By similarity). Activated through tyrosine phosphorylation by BMX. Tyrosine phosphorylated in response to IL6, IL11, LIF, CNTF, KITLG/SCF, CSF1, EGF, PDGF, IFN-alpha, LEP and OSM. Activated KIT promotes phosphorylation on tyrosine residues and subsequent translocation to the nucleus. Phosphorylated on serine upon DNA damage, probably by ATM or ATR. Serine phosphorylation is important for the formation of stable DNA-binding STAT3 homodimers and maximal transcriptional activity. ARL2BP may participate in keeping the phosphorylated state of STAT3 within the nucleus. Upon LPS challenge, phosphorylated within the nucleus by IRAK1. Upon erythropoietin treatment, phosphorylated on Ser-727 by RPS6KA5. Phosphorylation at Tyr-705 by PTK6 or FER leads to an increase of its transcriptional activity. Dephosphorylation on tyrosine residues by PTPN2 negatively regulates IL6/interleukin-6 signaling.

細胞内局在 Cytoplasm. Nucleus. Shuttles between the nucleus and the cytoplasm. Translocated into the nucleus upon tyrosine phosphorylation and dimerization, in response to signaling by activated FGFR1, FGFR2, FGFR3 or FGFR4. Constitutive nuclear presence is independent of tyrosine

phosphorylation. Predominantly present in the cytoplasm without stimuli. Upon leukemia inhibitory factor (LIF) stimulation, accumulates in the nucleus. The complex composed of BART and ARL2 plays an important role in the nuclear translocation and retention of STAT3. Identified in a complex with LYN and PAG1.

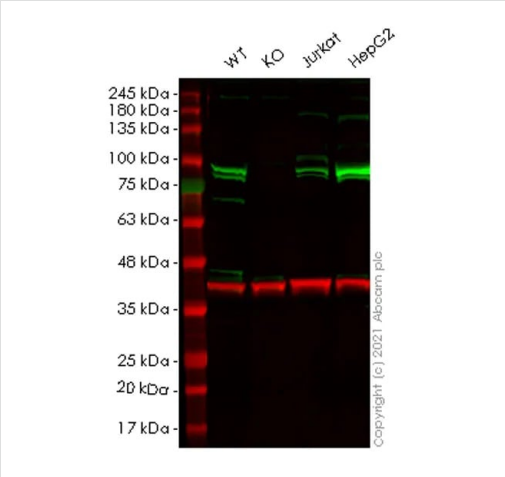
アプリケーション

The Abpromise guarantee **Abpromise保証は、次のテスト済みアプリケーションにおけるab263797の使用に適用されます**

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB		Use at an assay dependent concentration.

画像



Western blot - Human STAT3 knockout HeLa cell lysate (ab263797)

Blocking and diluting buffer and concentration: Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS.

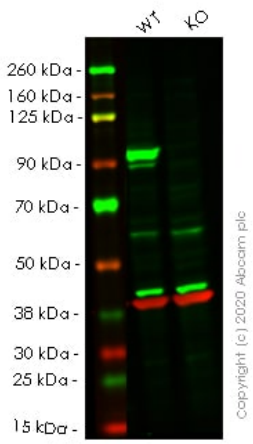
Lane 1: Wild-type HeLa (human cervix adenocarcinoma epithelial cell) serum starved overnight, then treated with 50 ng/ml IFN alpha for 30 minutes, whole cell lysate (20µg)
Lane 2: STAT3 knockout HeLa serum starved overnight, then treated with 50 ng/ml IFN alpha for 30 minutes, whole cell lysate (20µg)
Lane 3: Jurkat (human t cell leukemia cell line from peripheral blood) treated with 50 ng/ml IFN alpha for 30 minutes, whole cell lysate (20µg)
Lane 4: HepG2 (human hepatocellar carcinoma epithelial cell) serum starved overnight, then treated with 100 ng/ml IL-6 for 30 minutes, whole cell lysate (20µg)

Lanes 1-4: Merged signal (red and green). Green - **ab267373** observed at 88 kDa. Red - loading control **ab8245** (Mouse monoclonal [6C5] to GAPDH) observed at 36 kDa.

Lanes 1-2: **ab267373** Anti-STAT3 (phospho Y705) antibody [EPR23968-52] was shown to specifically react with STAT3 in wild-type serum starved and then IFN alpha treated HeLa cells. Loss of signal was observed when serum starved and then IFN alpha treated knockout cell line **ab255436** (knockout cell lysate ab263797) was used. Wild-type and STAT3 knockout samples were subjected to SDS-PAGE. **ab267373** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated at 4°C overnight at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L

(IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 10000 dilution for 1 hour at room temperature before imaging.

Lysates loaded onto lanes 3-4 were made freshly and used in WB immediately to minimize protein degradation.



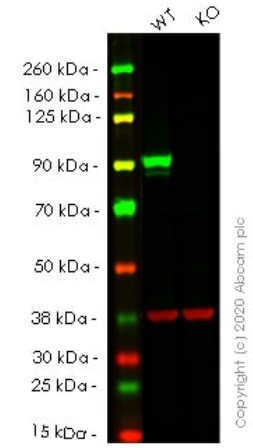
Western blot - Human STAT3 knockout HeLa cell lysate (ab263797)

Lane 1: Wild-type HeLa cell lysate (20µg)

Lane 2: STAT3 knockout HeLa cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - **ab68153** observed at 92 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab68153 Recombinant Anti-STAT3 antibody [EPR787Y] was shown to specifically react with STAT3 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab255436** (knockout cell lysate ab263797) was used. Wild-type and STAT3 knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab68153** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Human STAT3 knockout HeLa cell lysate (ab263797)

Lane 1: Wild-type HeLa cell lysate (20µg)

Lane 2: STAT3 knockout HeLa cell lysate (20µg)

Lanes 1- 2: Merged signal (red and green). Green - **ab109085** observed at 92 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab109085 Recombinant Anti-STAT3 antibody [EPR361] was shown to specifically react with STAT3 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab255436** (knockout cell lysate ab263797) was used. Wild-type and STAT3 knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1%

TBST with 3% non-fat dried milk. **ab109085** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Sanger Sequencing - Human STAT3 knockout HeLa cell lysate (ab263797)

Homozygous: 1 bp deletion in exon2

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