abcam

Product datasheet

STAT3 (Tyr705) In-Cell ELISA Kit ab126427

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Overview

Product name STAT3 (Tyr705) In-Cell ELISA Kit

Detection method Colorimetric
Sample type Adherent cells

Assay type Cell-based (qualitative)

Assay time 5h 10m

Assay duration Multiple steps standard assay

Species reactivity Reacts with: Mouse, Rat, Human

Product overview

ab126427 is a very rapid, convenient and sensitive assay kit that can monitor the activation or function of important biological pathways in cells. It can be used for measuring the relative amount of STAT3 (Tyr705) phosphorylation and screening the effects of various treatments, inhibitors (such as siRNA or chemicals), or activators in cultured human, mouse and rat cell lines. By determining STAT3 protein phosphorylation in your experimental model system, you can verify pathway activation in your cell lines without spending excess time and effort in preparing cell

lysate and performing an analysis of Western Blot.

In the STAT3 (Tyr705) In-Cell ELISA Kit, cells are seeded into a 96 well tissue culture plate. The cells are fixed after various treatments, inhibitors or activators. After blocking, Anti-Phospho-STAT3 (Tyr705) or Anti-STAT3 is pipetted into the wells and incubated. The wells are washed, and HRP-conjugated anti-mouse IgG is added to the wells. The wells are washed again, a TMB substrate solution is added to the wells and color develops in proportion to the amount of protein. The Stop Solution changes the color from blue to yellow, and the intensity of the color is measured

at 450 nm.

Platform Microplate

Properties

Storage instructions Store at -20°C. Please refer to protocols.

Components	1 x 96 tests
HRP-conjugated Anti-Mouse IgG Concentrate	1 x 10µl
Blocking Buffer Concentrate (5X)	1 x 20ml

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Components	1 x 96 tests
Fixing Solution	1 x 30ml
Uncoated 96-well Microplate	1 unit
Mouse Anti-Phospho-STAT3 (Tyr705) Concentrate (Item G)	1 x 10µl
Mouse Anti-STAT3 Concentrate (Item H)	1 x 10µl
Quenching Buffer Concentrate (30x)	1 x 2ml
Stop Solution	1 x 14ml
TMB One-Step Substrate Reagent	1 x 12ml
Wash Buffer A Concentrate (20X)	1 x 30ml
Wash Buffer B Concentrate (20X)	1 x 30ml

Function

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.

Tissue specificity

Involvement in disease

Sequence similarities

Post-translational modifications

Heart, brain, placenta, lung, liver, skeletal muscle, kidney and pancreas.

 $\label{prop:continuous} \mbox{Hyperimmunoglobulin E recurrent infection syndrome, autosomal dominant}$

Autoimmune disease, multisystem, infantile-onset

Belongs to the transcription factor STAT family.

Contains 1 SH2 domain.

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

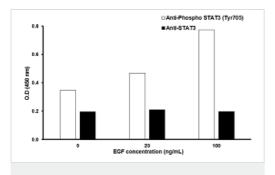
Phosphorylated on serine upon DNA damage, probably by ATM or ATR. Serine phosphorylation is important for the formation of stable DNA-binding STAT3 homodimers and maximal transcriptional activity. ARL2BP may participate in keeping the phosphorylated state of STAT3 within the nucleus. Upon LPS challenge, 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.

Cellular localization

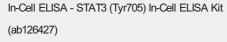
Cytoplasm. Nucleus. Shuttles between the nucleus and the cytoplasm. Translocated into the nucleus upon tyrosine phosphorylation and dimerization, in response to signaling by activated FGFR1, FGFR2, FGFR3 or FGFR4. Constitutive nuclear presence is independent of tyrosine

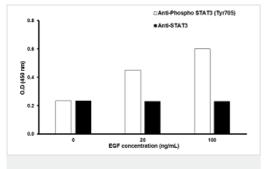
phosphorylation. Predominantly present in the cytoplasm without stimuli. Upon leukemia inhibitory factor (LIF) stimulation, accumulates in the nucleus. The complex composed of BART and ARL2 plays an important role in the nuclear translocation and retention of STAT3. Identified in a complex with LYN and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

Images



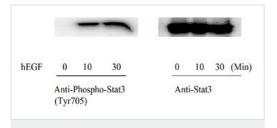
A431 cells were stimulated by different concentrations of EGF for 30 minutes at 37°C.





A431 cells were stimulated by different concentrations of EGF for 10 minutes at 37°C.

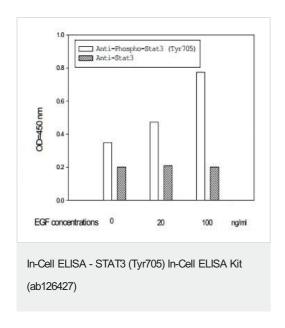
In-Cell ELISA - STAT3 (Tyr705) In-Cell ELISA Kit (ab126427)



A431 cells. Phospho-Stat3 (Tyr705) and Anti-Stat3 antibodies were used in both detection assays.

Western blot analysis of extracts from 100 ng/ml hEGF treated

Western blot - STAT3 (Tyr705) In-Cell ELISA Kit (ab126427)



A431 cells were stimulated by different concentrations of EGF for 30 minutes at 37°C.

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