abcam

Product datasheet

# p38 MAPK alpha (pT180/Y182 + Total) ELISA Kit ab221013

SimpleStep ELISA®

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## 概述

**产品名称** p38 MAPK alpha (pT180/Y182 + Total) ELISA试剂盒

**检测方法** Colorimetric

**精确度**

批次内

| 样品 | n | Mean | SD | CV% |
|---|---|---|---|---|
| (pT180/Y182) | 6 | | | 4.6% |
| (Total) | 6 | | | 9.2% |

批次间

| 样品 | n | Mean | SD | CV% |
|---|---|---|---|---|
| (pT180/Y182) | 3 | | | 2.5% |
| (Total) | 3 | | | 6.4% |

**样品类型** Cell Lysate, Tissue Homogenate

**检测类型** Semi-quantitative

**检测时间** 1h 30m

**实验步骤** One step assay

**种属反应性** **与反应:** Mouse, Human

**预测可用于:** Rat

**产品概述** p38 MAPK alpha (pT180/Y182 + Total) ELISA Kit (**ab176664**) has been re-developed with new capture and detector antibodies. This new kit has the same name but a different product number (ab221013). We have identified new recombinant monoclonal antibodies to use in the SimpleStep ELISA platform that provide a higher sensitivity when quantifying Total p38 MAPKα protein in human and mouse cells.

Abcam's p38 MAPKα (pT180/Y182) and p38 MAPKα (Total) *in vitro* SimpleStep ELISA™ (Enzyme-Linked Immunosorbent Assay) kit is designed for the semi-quantitative measurement of p38 MAPKα (pT180/Y182) and Total p38 MAPKα protein in human and mouse cells.

The SimpleStep ELISA™ employs an affinity tag labeled capture antibody and a reporter conjugated detector antibody which immunocapture the sample analyte in solution. This entire complex (capture antibody/analyte/detector antibody) is in turn immobilized via immunoaffinity of an anti-tag antibody coating the well. To perform the assay, samples or standards are added to the wells, followed by the antibody mix. After incubation, the wells are washed to remove unbound material. TMB substrate is added and during incubation is catalyzed by HRP, generating blue coloration. This reaction is then stopped by addition of Stop Solution completing any color change from blue to yellow. Signal is generated proportionally to the amount of bound analyte and the intensity is measured at 450 nm. Optionally, instead of the endpoint reading, development of TMB can be recorded kinetically at 600 nm.

**Sensitivity:**

p38 MAPK (pT180/Y182) = 0.2 ng/mL

p38 MAPK (Total) = 0.3 ng/mL

**As of May 2020, this kit was reformulated with new antibodies to maintain continued long term supply.**

**说明**

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.

**平台**

Microplate

## 性能

**存放说明**

Store at +4°C. Please refer to protocols.

| 组件 | 1 x 96 tests | 1 x 96 tests |
|---|---|---|
| 10X Wash Buffer PT | 1 x 20ml | 1 x 20ml |
| 50X Cell Extraction Enhancer Solution | 1 x 1ml | 1 x 1ml |
| 5X Cell Extraction Buffer PTR | 1 x 12ml | 1 x 12ml |
| Lyophilized p38 MAPK alpha Control Lysate | 1 vial | 1 vial |
| p38 MAPK alpha (pT180/Y182) Capture Antibody | 1 x 1.5ml | 1 x 1.5ml |
| p38 MAPK alpha (pT180/Y182) Detector Antibody | 1 x 1.5ml | 1 x 1.5ml |
| p38 MAPK alpha (Total) Capture Antibody | 1 x 1.5ml | 1 x 1.5ml |
| p38 MAPK alpha (Total) Detector Antibody | 1 x 1.5ml | 1 x 1.5ml |
| Plate Seal | 1 unit | 1 unit |

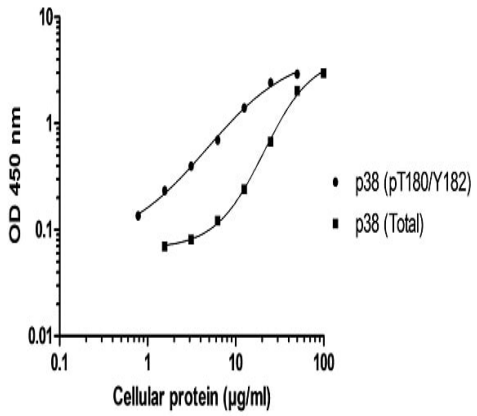
| 组件 | 1 x 96 tests | 1 x 96 tests |
|---|---|---|
| SimpleStep Pre-Coated 96-Well Microplate (ab206978) | 1 unit | 1 unit |
| Stop Solution | 1 x 12ml | 1 x 12ml |
| TMB Substrate | 1 x 12ml | 1 x 12ml |

**功能**

Serine/threonine kinase which acts as an essential component of the MAP kinase signal transduction pathway. MAPK14 is one of the four p38 MAPKs which play an important role in the cascades of cellular responses evoked by extracellular stimuli such as proinflammatory cytokines or physical stress leading to direct activation of transcription factors. Accordingly, p38 MAPKs phosphorylate a broad range of proteins and it has been estimated that they may have approximately 200 to 300 substrates each. Some of the targets are downstream kinases which are activated through phosphorylation and further phosphorylate additionnal targets. RPS6KA5/MSK1 and RPS6KA4/MSK2 can directly phosphorylate and activate transcription factors such as CREB1, ATF1, the NF-kappa-B isoform RELA/NFKB3, STAT1 and STAT3, but can also phosphorylate histone H3 and the nucleosomal protein HMGN1. RPS6KA5/MSK1 and RPS6KA4/MSK2 play important roles in the rapid induction of immediate-early genes in response to stress or mitogenic stimuli, either by inducing chromatin remodeling or by recruiting the transcription machinery. On the other hand, two other kinase targets, MAPKAPK2/MK2 and MAPKAPK3/MK3, participate in the control of gene expression mostly at the post-transcriptional level, by phosphorylating ZFP36 (tristetraprolin) and ELAVL1, and by regulating EEF2K, which is important for the elongation of mRNA during translation. MKNK1/MNK1 and MKNK2/MNK2, two other kinases activated by p38 MAPKs, regulate protein synthesis by phosphorylating the initiation factor EIF4E2. MAPK14 interacts also with casein kinase II, leading to its activation through autophosphorylation and further phosphorylation of TP53/p53. In the cytoplasm, the p38 MAPK pathway is an important regulator of protein turnover. For example, CFLAR is an inhibitor of TNF-induced apoptosis whose proteasome-mediated degradation is regulated by p38 MAPK phosphorylation. In a similar way, MAPK14 phosphorylates the ubiquitin ligase SIAH2, regulating its activity towards EGLN3. MAPK14 may also inhibit the lysosomal degradation pathway of autophagy by interfering with the intracellular trafficking of the transmembrane protein ATG9. Another function of MAPK14 is to regulate the endocytosis of membrane receptors by different mechanisms that impinge on the small GTPase RAB5A. In addition, clathrin-mediated EGFR internalization induced by inflammatory cytokines and UV irradiation depends on MAPK14-mediated phosphorylation of EGFR itself as well as of RAB5A effectors. Ectodomain shedding of transmembrane proteins is regulated by p38 MAPKs as well. In response to inflammatory stimuli, p38 MAPKs phosphorylate the membrane-associated metalloprotease ADAM17. Such phosphorylation is required for ADAM17-mediated ectodomain shedding of TGF-alpha family ligands, which results in the activation of EGFR signaling and cell proliferation. Another p38 MAPK substrate is FGFR1. FGFR1 can be translocated from the extracellular space into the cytosol and nucleus of target cells, and regulates processes such as rRNA synthesis and cell growth. FGFR1 translocation requires p38 MAPK activation. In the nucleus, many transcription factors are phosphorylated and activated by p38 MAPKs in response to different stimuli. Classical examples include ATF1, ATF2, ATF6, ELK1, PTPRH, DDIT3, TP53/p53 and MEF2C and MEF2A. The p38 MAPKs are emerging as important modulators of gene expression by regulating chromatin modifiers and remodelers. The promoters of several genes involved in the inflammatory response, such as IL6, IL8 and IL12B, display a p38 MAPK-dependent enrichment of histone H3 phosphorylation on 'Ser-10' (H3S10ph) in LPS-stimulated myeloid cells. This phosphorylation enhances the accessibility of the cryptic NF-kappa-B-binding sites marking

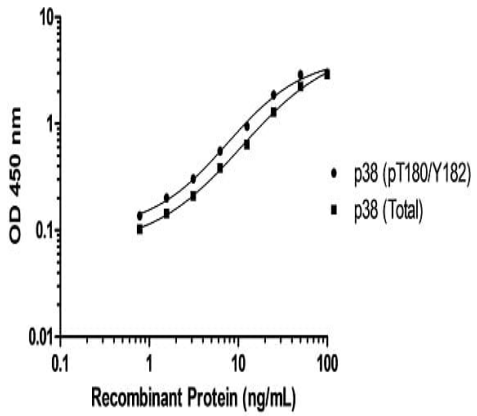
| | |
|---|---|
| | promoters for increased NF-kappa-B recruitment. Phosphorylates CDC25B and CDC25C which is required for binding to 14-3-3 proteins and leads to initiation of a G2 delay after ultraviolet radiation. Phosphorylates TIAR following DNA damage, releasing TIAR from GADD45A mRNA and preventing mRNA degradation. The p38 MAPKs may also have kinase-independent roles, which are thought to be due to the binding to targets in the absence of phosphorylation. Protein O-Glc-N-acylation catalyzed by the OGT is regulated by MAPK14, and, although OGT does not seem to be phosphorylated by MAPK14, their interaction increases upon MAPK14 activation induced by glucose deprivation. This interaction may regulate OGT activity by recruiting it to specific targets such as neurofilament H, stimulating its O-Glc-N-acylation. Required in mid-fetal development for the growth of embryo-derived blood vessels in the labyrinth layer of the placenta. Also plays an essential role in developmental and stress-induced erythropoiesis, through regulation of EPO gene expression. Isoform MXI2 activation is stimulated by mitogens and oxidative stress and only poorly phosphorylates ELK1 and ATF2. Isoform EXIP may play a role in the early onset of apoptosis. |
| **组织特异性** | Brain, heart, placenta, pancreas and skeletal muscle. Expressed to a lesser extent in lung, liver and kidney. |
| **序列相似性** | Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. MAP kinase subfamily.<br>Contains 1 protein kinase domain. |
| **结构域** | The TXY motif contains the threonine and tyrosine residues whose phosphorylation activates the MAP kinases. |
| **翻译后修饰** | Dually phosphorylated on Thr-180 and Tyr-182 by the MAP2Ks MAP2K3/MKK3, MAP2K4/MKK4 and MAP2K6/MKK6 in response to inflammatory citokines, environmental stress or growth factors, which a ctivates the enzyme. Dual phosphorylation can also be mediated by TAB1-mediated autophosphorylation. TCR engagement in T-cells also leads to Tyr-323 phosphorylation by ZAP70. Dephosphorylated and inactivated by DUPS1, DUSP10 and DUSP16.<br>Acetylated at Lys-53 and Lys-152 by KAT2B and EP300. Acetylation at Lys-53 increases the affinity for ATP and enhances kinase activity. Lys-53 and Lys-152 are deacetylated by HDAC3.<br>Ubiquitinated. Ubiquitination leads to degradation by the proteasome pathway. |
| **细胞定位** | Cytoplasm. Nucleus. |

**图片**



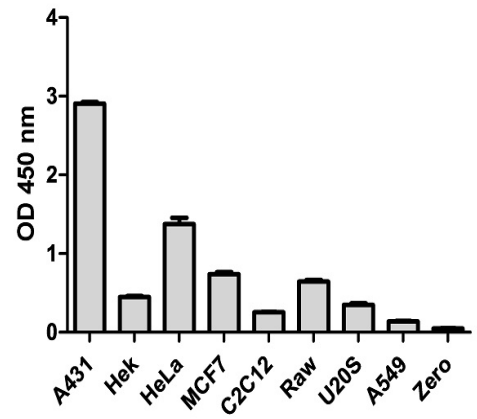

Example of p38 MAPKa (pT180/Y182) and p38 MAPKa (Total) cell lysate standard curve

Raw data values are fitted to a four-parameter, variable slope curve (+/- SD).



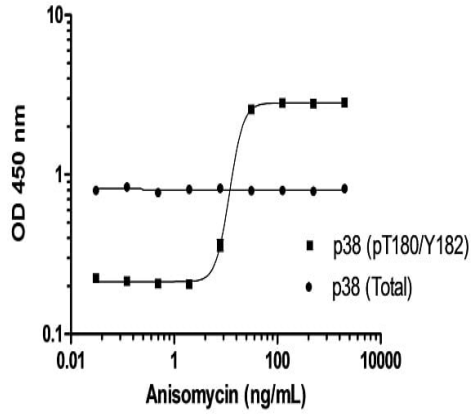

Example of p38 MAPKa (pT180/Y182) and p38 MAPKa (Total) recombinant protein standard curve

Raw data values are fitted to a four-parameter, variable slope curve (+/- SD).



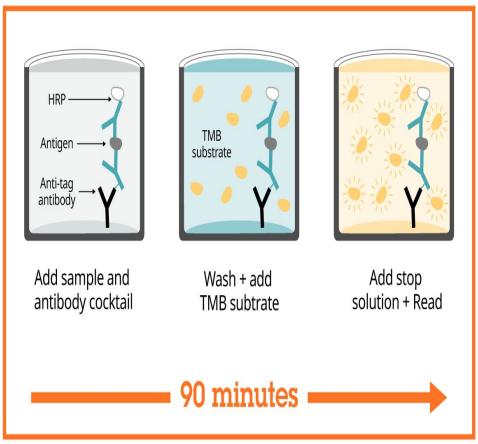

Cell line analysis for Total p38 MAPKa from 100 µg/mL preparations of cell extracts

Data from triplicate measurements (mean +/- SD) are plotted and compared to 1X Cell Extraction Buffer PTR (zero).



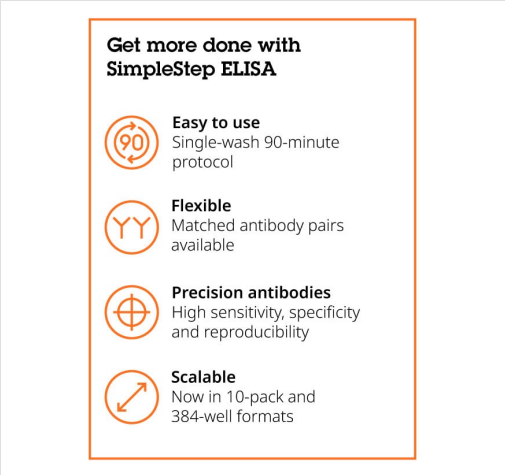

Induction of p38 MAPKa (pT180/Y182) phosphorylation in HeLa cells in response to anisomycin treatment

HeLa cells were cultured in 96-well tissue culture plates and treated (15 min) with a dose-range of anisomycin before cell lysis. Data from quadruplicate measurements of p38 MAPKα (pT180/Y182) are plotted and compared against Total p38 MAPKα protein levels.




Sandwich ELISA - p38 MAPK alpha (pT180/Y182 + Total) ELISA Kit (ab221013)

SimpleStep ELISA technology allows the formation of the antibody-antigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.




Sandwich ELISA - p38 MAPK alpha (pT180/Y182 + Total) ELISA Kit (ab221013)

To learn more about the advantages of SimpleStep ELISA® kits see **here**.

**Please note:** All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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