# abcam

# Product datasheet

# JNK 1/2 (pT183/Y185) ELISA Kit ab176645

SimpleStep ELISA

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#### Overview

Product name JNK 1/2 (pT183/Y185) ELISA Kit

**Detection method** Colorimetric

Precision Intra-assay

Sample	n	Mean	SD	CV%
HEK extract	6			2.7%

Inter-assay

Sample	n	Mean	SD	CV%	
HEK extract	3			3.8%	

Sample type Cell Lysate, Tissue Homogenate

**Assay type** Semi-quantitative

**Sensitivity** 0.5 ng/ml

Range 1 ng/ml - 100 ng/ml

Assay time 1h 30m

**Assay duration** One step assay

Species reactivity Reacts with: Mouse, Human

Predicted to work with: Rat

**Product overview** Abcam's JNK1/2 (pT183/Y185) *in vitro* SimpleStep ELISA™ (Enzyme-Linked Immunosorbent

Assay) kit is designed for the semi-quantitative measurement of JNK1/2 (pT183/Y185) 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

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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.

# As of October 2019, this kit was reformulated with new antibodies to maintain continued long term supply.

**Notes** 

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.

**Platform** Microplate

#### **Properties**

#### Storage instructions

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

Components	1 x 96 tests	1 x 96 tests
10X Wash Buffer PT	1 x 15ml	1 x 15ml
50X Cell Extraction Enhancer Solution	1 x 1ml	1 x 1ml
5X Cell Extraction Buffer PTR	1 x 12ml	1 x 12ml
JNK1/2 (pT183/Y185) Capture Antibody	1 x 3ml	1 x 3ml
JNK1/2 (pT183/Y185) Detector Antibody	1 x 3ml	1 x 3ml
Lyophilized JNK1/2 Control Lysate	1 vial	1 vial
Plate Seal	1 unit	1 unit
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

#### **Function**

Responds to activation by environmental stress and pro-inflammatory cytokines by phosphorylating a number of transcription factors, primarily components of AP-1 such as JUN, JDP2 and ATF2 and thus regulates AP-1 transcriptional activity. In T-cells, JNK1 and JNK2 are required for polarized differentiation of T-helper cells into Th1 cells (By similarity). Phosphorylates heat shock factor protein 4 (HSF4).

JNK1 isoforms display different binding patterns: beta-1 preferentially binds to c-Jun, whereas alpha-1, alpha-2, and beta-2 have a similar low level of binding to both c-Jun or ATF2. However, there is no correlation between binding and phosphorylation, which is achieved at about the same efficiency by all isoforms.

### Sequence similarities

Belongs to the protein kinase superfamily. CMGC Ser/Thr protein kinase family. MAP kinase subfamily.

Contains 1 protein kinase domain.

**Domain** 

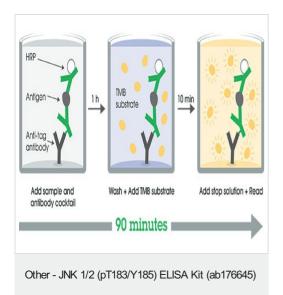
The TXY motif contains the threonine and tyrosine residues whose phosphorylation activates the MAP kinases.

Post-translational

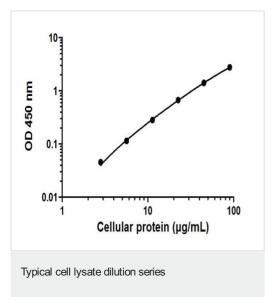
modifications

Dually phosphorylated on Thr-183 and Tyr-185, which activates the enzyme.

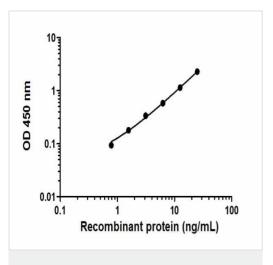
#### **Images**



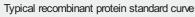
SimpleStep ELISA technology allows the formation of the antibodyantigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.

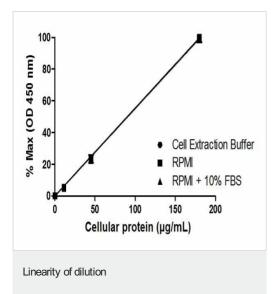


Example of a typical JNK1/2 (pT183/Y185) cell lysate dilution series. Background-subtracted data values (mean +/- SD) are graphed.

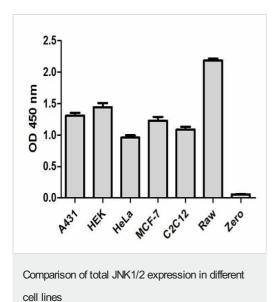


Example of a typical JNK1/2 (pT183/Y185) recombinant protein standard curve. The proportion of total protein that is phosphorylated is unknown - data is indicative only. Background-subtracted data values (mean +/- SD) are graphed.

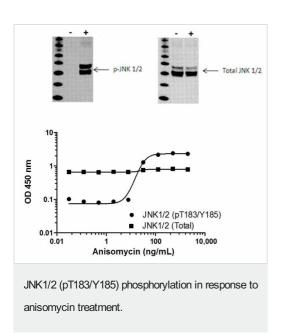




Linearity of dilution in representative sample matrices. Cellular lysates were prepared at 3 concentrations in common media containing 1 x Cell Extraction Buffer PTR. Data from duplicate measurements of JNK1/2 (pT183/Y185) are normalized and plotted.



Cell line analysis for Total JNK1/2 from 200  $\mu$ g/mL preparations of cell extracts. Data from triplicate measurements (mean +/- SD) are plotted and compared to 1X Cell Extraction Buffer PTR (zero).



Induction of JNK1/2 (pT183/Y185) phosphorylation in HeLa cells in response to anisomycin treatment. HeLa cells were cultured in 96-well tissue culture plates and treated (30 min) with a dose-range of anisomycin before cell lysis. Data from quadruplicate measurements of JNK1/2 (pT183/Y185) are plotted and compared against Total JNK1/2 protein levels. Comparative JNK1/2 (pT183/Y185) and JNK1/2 (Total) data also shown by Western Blot.

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