

ERK1 (phospho T202 + Y204) + ERK2 (phospho T185 + Y187) + Total ELISA Kit ab176660

SimpleStep ELISA

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Overview

Product name ERK1 (phospho T202 + Y204) + ERK2 (phospho T185 + Y187) + Total ELISA Kit

Detection method Colorimetric

Precision Intra-assay

Sample	n	Mean	SD	CV%
(pT202/Y204)	6			3.3%
(Total)	6			3%

Inter-assay

Sample	n	Mean	SD	CV%
(pT202/Y204)	3			1.3%
(Total)	3			4.9%

Sample type Cell Lysate, Tissue Homogenate

Assay type Semi-quantitative

Assay time 1h 30m

Assay duration One step assay

Species reactivity Reacts with: Mouse, Human

Predicted to work with: Rat 

Product overview Abcam's ERK1 (phospho T202 + Y204) + ERK2 (phospho T185 + Y187) + Total ELISA kit is designed for the semi-quantitative measurement of ERK1/2 (pT202/Y204) and Total ERK1/2 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.

## ASSAY SPECIFICITY

The ERK1/2 (pT202/Y204) assay detects endogenous levels of ERK1/2 (GenBank Accessions NP 002737.2 [ERK1], NP 620407 [ERK2]) in cellular lysates, only when phosphorylated at Thr202/Tyr204.

The ERK1/2 Total assay detects endogenous levels of ERK1/2 (GenBank Accessions NP 002737.2 [ERK1], NP 620407 [ERK2]) in cellular lysates, irrespective of phosphorylation status.

## SPECIES REACTIVITY

This kit detects ERK1/2 (pT202/Y204) and ERK1/2 Total in Human and mouse cell culture extracts. Detection in rat samples is also expected. Other species should be tested on a case-by-case basis.

Serum and plasma samples have not been tested with this kit.

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

Notes	<b>Estimated sensitivity:</b> Phospho-ERK (Thr202/Tyr204): 0.1 ng/mL (tested with recombinant protein), Total ERK1/2: 0.6 ng/mL (tested with recombinant protein)
	<b>Range:</b> Phospho-ERK (Thr202/Tyr204): 0.2-20ng/mL, Total ERK1/2: 0.6-60 ng/mL
	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.

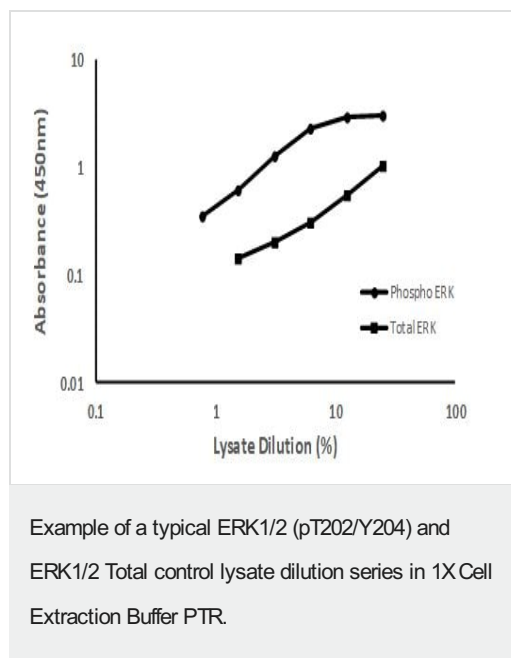
Components	1 x 96 tests	1 x 96 tests
10X Wash Buffer PT	1 x 15ml	1 x 15ml

Components	1 x 96 tests	1 x 96 tests
50X Cell Extraction Enhancer Solution	1 x 1ml	1 x 1ml
5X Cell Extraction Buffer PTR	1 x 10ml	1 x 10ml
ERK1/2 (pT202/Y204) Capture Antibody	1 x 1.5ml	1 x 1.5ml
ERK1/2 (pT202/Y204) Detector Antibody	1 x 1.5ml	1 x 1.5ml
ERK1/2 (Total) Capture Antibody	1 x 1.5ml	1 x 1.5ml
ERK1/2 (Total) Detector Antibody	1 x 1.5ml	1 x 1.5ml
Lyophilized ERK1/2 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

## Cellular localization

ERK2: Nucleus.

## Images



The ERK1/2 lysate dilution series was prepared as described.

Raw data values are shown in the table.

Kit Control lysates are provided at a concentration that give consistent signal between different lots. Lysates are produced and formulated by signal intensity to be consistent to within 30% of the previous lot. As such, Control lysates are not provided with a protein concentration.

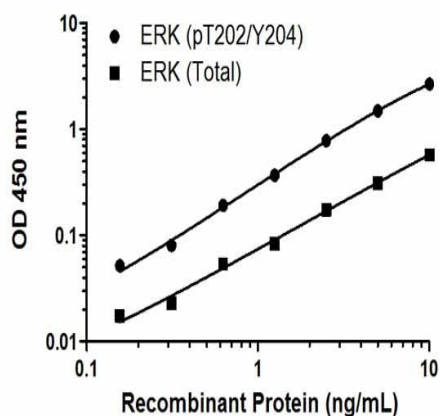
Lysate Dilution Series Measurements						
Control Lysate (%)	ERK 1/2 (pT202/Y204)			ERK 1/2 Total		
	O.D 450 nm		Mean O.D	O.D 450 nm		Mean O.D
	1	2		1	2	
0.000	0.046	0.050	0.048	0.091	0.100	0.096
0.78	0.343	0.356	0.350	-	-	-
1.56	0.593	0.631	0.612	0.137	0.150	0.144
3.13	1.231	1.273	1.252	0.198	0.204	0.201
6.25	2.283	2.222	2.253	0.291	0.313	0.302
12.5	2.936	2.866	2.901	0.545	0.542	0.544
25	3.040	3.024	3.032	1.011	1.014	1.013

Example of a typical ERK1/2 (pT202/Y204) and ERK1/2 Total control lysate dilution series in 1X Cell Extraction Buffer PTR.

The ERK1/2 lysate dilution series was prepared as described.

Raw data values are shown in the table.

Kit Control lysates are provided at a concentration that give consistent signal between different lots. Lysates are produced and formulated by signal intensity to be consistent to within 30% of the previous lot. As such, Control lysates are not provided with a protein concentration.



Typical recombinant protein standard curve

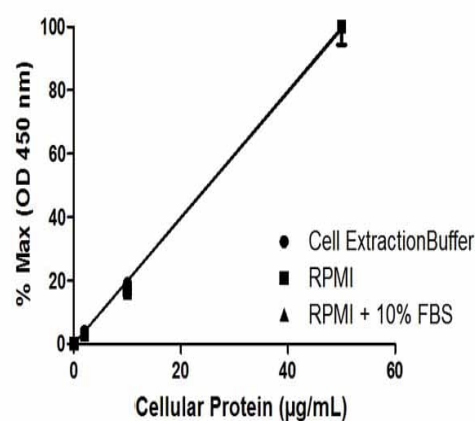
Example of a typical ERK1/2 (pT202/Y204) and ERK1/2 (Total) recombinant protein standard curve. The proportion of total protein that is phosphorylated is unknown - data is indicative only.

Background-subtracted data values (mean +/- SD) are graphed.

Recombinant Protein Standard Curve Measurements						
Concentration (ng/mL)	ERK1/2 (p-T202/Y204)			ERK1/2 Total		
	O.D 450 nm		Mean O.D	O.D 450 nm		Mean O.D
	1	2		1	2	
0.000	0.047	0.046	0.047	0.072	0.072	0.072
0.156	0.102	0.095	0.099	0.090	0.089	0.090
0.313	0.126	0.128	0.127	0.094	0.096	0.095
0.625	0.244	0.233	0.239	0.127	0.125	0.126
1.250	0.413	0.415	0.414	0.163	0.149	0.156
2.500	0.844	0.811	0.828	0.264	0.231	0.248
5.000	1.506	1.573	1.540	0.375	0.394	0.385
10.00	2.727	2.707	2.717	0.626	0.668	0.647

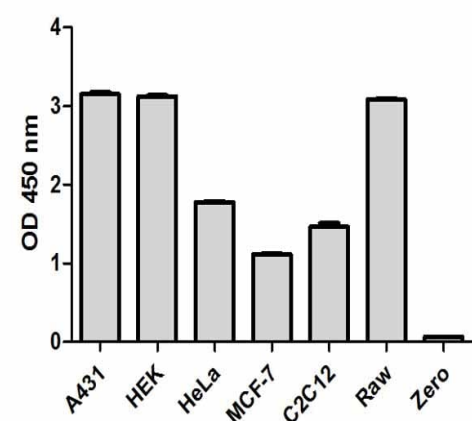
Example of a typical ERK1/2 (pT202/Y204) and ERK1/2 Total recombinant protein standard curve.

Raw data values are shown in the table. Background-subtracted data values (mean +/- SD) are graphed.



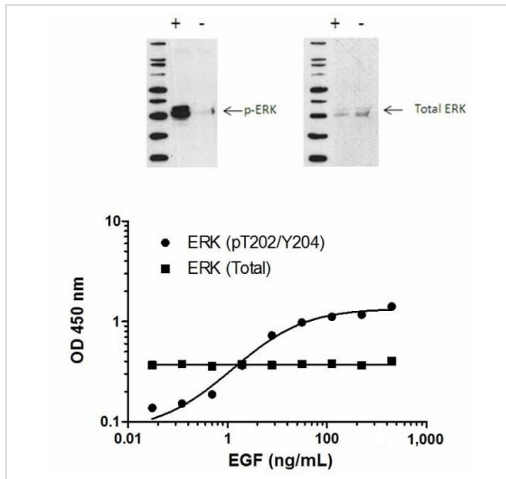
Linearity of dilution in representative sample matrices. Cellular lysates were prepared at 3 concentrations in common media containing 1X Cell Extraction Buffer PTR. Data from duplicate measurements of ERK1/2 (pT202/Y204) are normalized and plotted.

Linearity of dilution



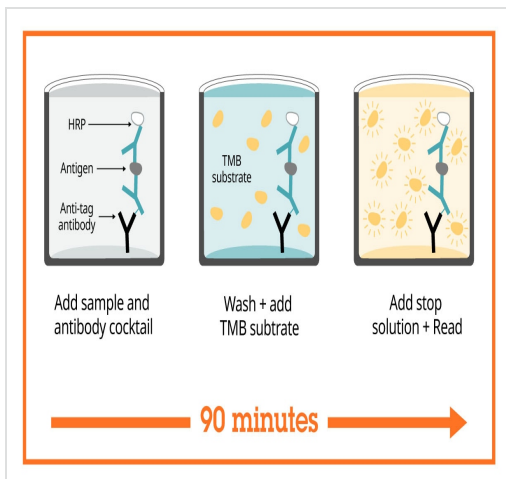
Cell line analysis for Total ERK1/2 from 100 µg/mL preparations of cell extracts. Data from triplicate measurements (mean +/- SD) are plotted and compared to 1X Cell Extraction Buffer PTR (zero).

Comparison of ERK1/2 expression in different cell lines



ERK1/2 (pT202/Y204) phosphorylation in response to EGF treatment

Induction of ERK1/2 (pT202/Y204) phosphorylation in MCF-7 cells in response to EGF treatment. MCF-7 cells were cultured in 96-well tissue culture plates, serum starved and treated (10 min) with a dose-range of EGF before cell lysis. Data from quadruplicate measurements of ERK1/2 (pT202/Y204) are plotted and compared against Total ERK1/2 protein levels. Comparative ERK1/2 (pT202/Y204) and ERK1/2 (Total) data also shown by Western Blot.



Sandwich ELISA - ERK1 (phospho T202 + Y204) + ERK2 (phospho T185 + Y187) + Total ELISA Kit (ab176660)

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.

**Get more done with  
SimpleStep ELISA**



**Easy to use**  
Single-wash 90-minute  
protocol



**Flexible**  
Matched antibody pairs  
available



**Precision antibodies**  
High sensitivity, specificity  
and reproducibility



**Scalable**  
Now in 10-pack and  
384-well formats

To learn more about the advantages of SimpleStep ELISA® kits  
see [here](#).

Sandwich ELISA - ERK1 (phospho T202 + Y204) +  
ERK2 (phospho T185 + Y187) + Total ELISA Kit  
(ab176660)

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

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