abcam

Product datasheet

Human AKT3 (pS473) ELISA Kit ab270887

Recombinant SimpleStep ELISA

5 Images

Overview

Product name Human AKT3 (pS473) ELISA Kit

Detection method Colorimetric

Precision

Intra-assay

Sample	n	Mean	SD	CV%
Extract	3			2.5%

Inter-assay

Sample	n	Mean	SD	CV%
Extract	24			3.4%

Sample type Cell Lysate

Assay type Sandwich (quantitative)

Sensitivity 19.65 pg/ml

Range 46.88 pg/ml - 3000 pg/ml

Recovery

Sample specific recovery

Sample type	Average %	Range
Cell Lysate	97	94% - 107%

Assay time 1h 30m

Assay duration One step assay

Species reactivity **Reacts with:** Human

Product overview Human ATK (pS473) ELISA kit (ab270887) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of ATK (pS473) protein in human cell extracts. It uses our proprietary SimpleStep ELISA® technology. Quantitate human ATK (pS473) with 5.9 units/mL sensitivity.

SimpleStep ELISA® technology employs capture antibodies conjugated to an affinity tag that is

recognized by the monoclonal antibody used to coat our SimpleStep ELISA® plates. This approach to sandwich ELISA allows the formation of the antibody-analyte sandwich complex in a single step, significantly reducing assay time. See the SimpleStep ELISA® protocol summary in the image section for further details. Our SimpleStep ELISA® technology provides several benefits:

-Single-wash protocol reduces assay time to 90 minutes or less
-High sensitivity, specificity and reproducibility from superior antibodies
-Fully validated in biological samples
-96-wells plate breakable into 12 x 8 wells strips

A 384-well SimpleStep ELISA® microplate (**ab203359**) is available to use as an alternative to the 96-well microplate provided with SimpleStep ELISA® kits.

Notes

AKT3 is a member of the AKT family that includes AKT1 and AKT2. These serine/threonine-protein kinases are involved in diverse processes including glucose uptake, cell cycle progression, and apoptosis. AKT3 plays an important role in brain development and is crucial for glioma cells malignancy. Phosphorylation of Serine 473 is a known regulator of AKT's kinase activity. The antibodies for this kit were raised against an AKT3-specific N-terminal fragment, as well as a phospho-peptide encompassing the 473-serine site, which shares high sequence identity for all three AKT members. The protein standard in this product is recombinant human AKT3 ligated with peptide containing phosphorylated Serine 416.

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

Pre-coated microplate (12 x 8 well strips)

Properties

Storage instructions

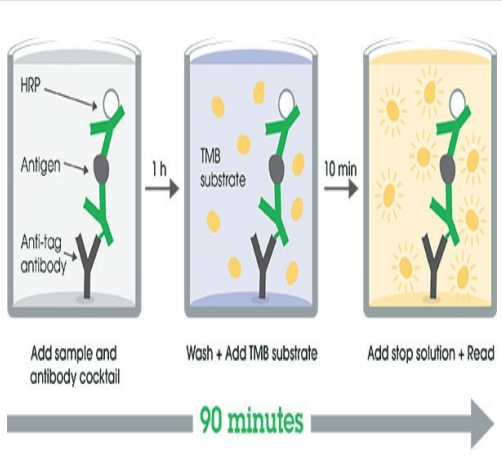
Store at +4°C. Please refer to protocols.

Components	1 x 96 tests
10X Human AKT3 (pS473) Capture Antibody	1 x 600µl
10X Human AKT3 (pS473) Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
5X Cell Extraction Buffer PTR (ab193970)	1 x 10ml
Antibody Diluent CPI - HAMA Blocker (ab193969)	1 x 6ml
Human AKT3 (pS473) Lyophilized Recombinant Protein	2 vials
Plate Seals	1 unit
Sample Diluent NS (ab193972)	1 x 12ml

Components	1 x 96 tests
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit
Stop Solution	1 x 12ml
TMB Development Solution	1 x 12ml

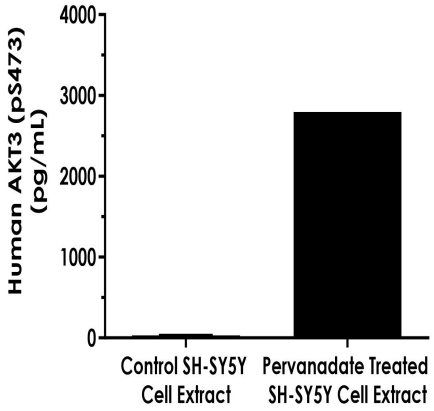
Function	IGF-1 leads to the activation of AKT3, which may play a role in regulating cell survival. Capable of phosphorylating several known proteins. Truncated isoform 2/PKB gamma 1 without the second serine phosphorylation site could still be stimulated but to a lesser extent.
Tissue specificity	In adult tissues, it is highly expressed in brain, lung and kidney, but weakly in heart, testis and liver. In fetal tissues, it is highly expressed in heart, liver and brain and not at all in kidney.
Sequence similarities	Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family. RAC subfamily. Contains 1 AGC-kinase C-terminal domain. Contains 1 PH domain. Contains 1 protein kinase domain.
Domain	Binding of the PH domain to the phosphatidylinositol 3-kinase alpha (PI(3)K) results in its targeting to the plasma membrane.
Post-translational modifications	Phosphorylation on Thr-305 and Ser-472 is required for full activity (By similarity). Phosphorylated upon DNA damage, probably by ATM or ATR. Ubiquitinated. When fully phosphorylated and translocated into the nucleus, undergoes 'Lys-48'-polyubiquitination catalyzed by TTC3, leading to its degradation by the proteasome.
Cellular localization	Cytoplasm. Membrane. Membrane-associated after cell stimulation leading to its translocation.

Images



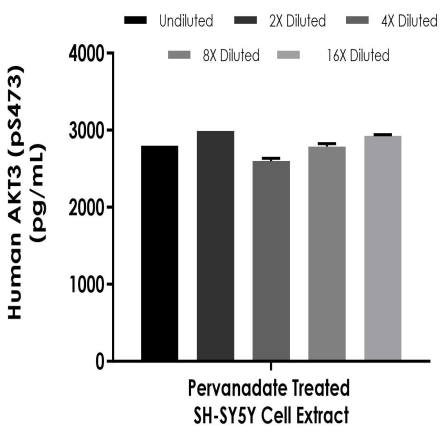
SimpleStep ELISA Protocol Diagram

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.



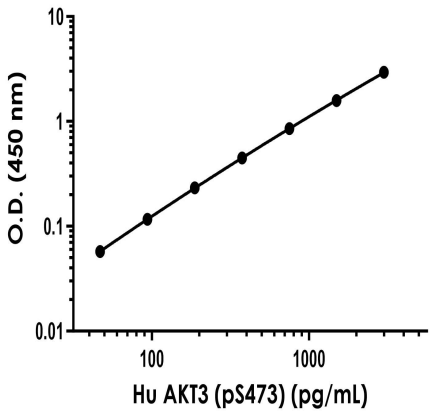
Interpolated concentrations of human AKT3 (pS473) in treated versus untreated SH-SY5Y cell extract

Comparison of native AKT3 (pS473) concentration in treated versus control cell extracts. SH-SY5Y cells were treated with 10 µM pervanadate for 20 minutes, scraped and buffer exchanged into PBS + 20 mM NaF, followed by protein extraction. Mock control SH-SY5Y cells accompanied cell treatment. Concentrations of AKT3 (pS473) were measured in duplicate and interpolated against the standard curve. Results show the mean AKT3 (pS473) concentration in control cell extract was 30 pg/mL compared to 2,795 pg/mL in pervanadate treated samples.



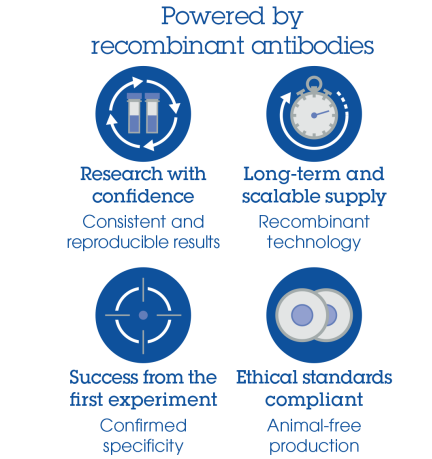
Interpolated concentrations of human AKT3 (pS473) in treated SH-SY5Y cell extract

Interpolated concentration of native AKT3 (pS473) was measured in duplicate at different sample concentrations. Undiluted samples are 0.5 mg/mL treated SH-SY5Y cell extract (10 µM pervanadate for 20 min). The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). Sample dilutions are made in 1X Cell Extraction Buffer PTR.



Example of human AKT3 (pS473) standard curve in 1X Cell Extraction Buffer PTR.

Example of human AKT3 (pS473) standard curve. Background-subtracted data values (mean +/- SD) are graphed.



Sandwich ELISA - Human AKT3 (pS473) ELISA Kit (ab270887)

To learn more about the advantages of recombinant antibodies see **here**.

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