

Product datasheet

Mouse PDK4 ELISA Kit ab215544

SimpleStep ELISA

[5 Images](#)

Overview

Product name Mouse PDK4 ELISA Kit

Detection method Colorimetric

Precision

Intra-assay

Sample	n	Mean	SD	CV%
Overall	5			2%

Inter-assay

Sample	n	Mean	SD	CV%
Overall	3			9%

Sample type Cell culture extracts, Tissue Extracts

Assay type Sandwich (quantitative)

Sensitivity 45 pg/ml

Range 0.234 ng/ml - 15 ng/ml

Recovery

Sample specific recovery

Sample type	Average %	Range
Cell culture extracts	98	94% - 103%
Mouse muscle extract	98	95% - 100%

Assay time 1h 30m

Assay duration One step assay

Species reactivity **Reacts with:** Mouse, Rat

Does not react with: Cow, Human

Product overview PDK4 *in vitro* SimpleStep ELISA (Enzyme-Linked Immunosorbent Assay) kit is designed for the quantitative measurement of PDK4 protein in mouse cell and tissue extract samples.

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.

Notes

PDK4 is a mitochondrial kinase that plays a key role in regulation of glucose and fatty acid metabolism and homeostasis via phosphorylation of the pyruvate dehydrogenase subunits PDHA1 and PDHA2. PDH phosphorylation inhibits pyruvate dehydrogenase activity, and thereby regulates metabolite flux through the tricarboxylic acid cycle, down-regulates aerobic respiration and inhibits the formation of acetyl-coenzyme A from pyruvate. Inhibition of pyruvate dehydrogenase decreases glucose utilization and increases fat metabolism in response to prolonged fasting and starvation. PDK4 plays an important role in maintaining normal blood glucose levels under starvation, and is involved in the insulin signaling cascade. Via its regulation of pyruvate dehydrogenase activity, PDK4 plays an important role in maintaining normal blood pH and in preventing the accumulation of ketone bodies under starvation. In the fed state, PDK4 mediates cellular responses to glucose levels and to a high-fat diet. PDK4 regulates both fatty acid oxidation and de novo fatty acid biosynthesis. PDK4 plays a role in the generation of reactive oxygen species.

Tested applications

Suitable for: Sandwich ELISA

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 Mouse PDK4 Capture Antibody	1 x 600µl
10X Mouse PDK4 Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
50X Cell Extraction Enhancer Solution (ab193971)	1 x 1ml
5X Cell Extraction Buffer PTR (ab193970)	1 x 10ml
Antibody Diluent 4BR	1 x 6ml
Mouse PDK4 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	Inhibits the mitochondrial pyruvate dehydrogenase complex by phosphorylation of the E1 alpha subunit, thus contributing to the regulation of glucose metabolism.
Tissue specificity	Ubiquitous; highest levels of expression in heart and skeletal muscle.
Sequence similarities	Belongs to the PDK/BCKDK protein kinase family. Contains 1 histidine kinase domain.
Cellular localization	Mitochondrion matrix.

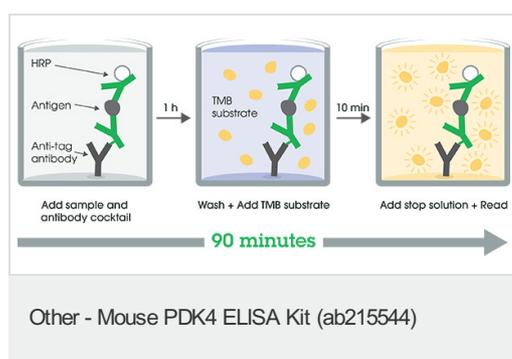
Applications

Our [Abpromise guarantee](#) covers the use of **ab215544** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Sandwich ELISA		Use at an assay dependent concentration.

Images

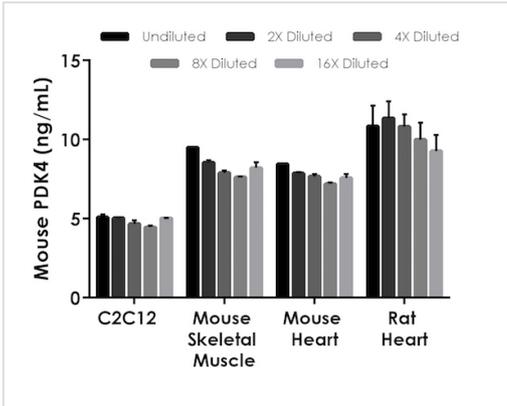


SimpleStep ELISA technology allows the formation of the antibody-antigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.



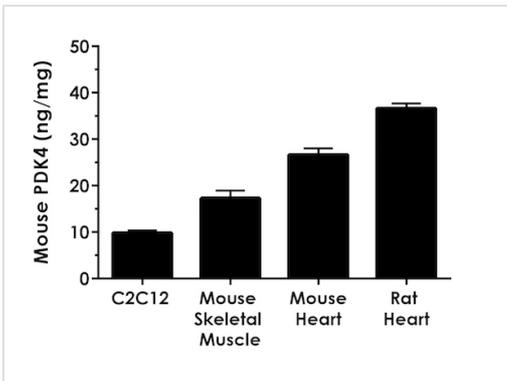
Example of mouse PDK4 standard curve.

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



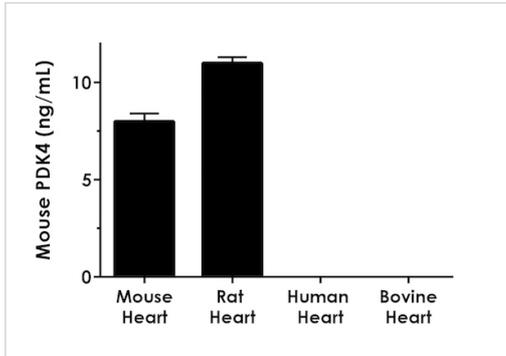
Interpolated concentrations of native PDK4 in C2C12 cell extract sample, mouse skeletal muscle tissue extract and mouse and rat heart tissue extracts.

Interpolated concentrations of native PDK4 in C2C12 cell extract sample based on a 500 µg/mL extract load, mouse skeletal muscle tissue extract based on a 500 µg/mL extract load, and mouse and rat heart tissue extract based on a 300 µg/mL extract load. The concentrations of PDK4 were measured in duplicate and interpolated from the PDK4 standard curve and corrected for sample dilution. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean PDK4 concentration was determined to be 4.8 ng/mL in C2C12 extract, 8.4 ng/mL in mouse skeletal muscle extract, 7.8 ng/mL in mouse heart extract, and 10.5 ng/mL in rat heart extract.



Interpolated concentrations of PDK4 in various cell and tissue extracts.

Interpolated concentrations of PDK4 in various cell and tissue extracts. The concentrations of PDK4 were measured in three different dilutions in duplicate and interpolated from the PDK4 standard curve and corrected for sample dilution. The interpolated dilution factor corrected values are plotted in ng PDK4 per mg of extract (mean +/- SD, n=3). PDK4 concentration was determined to be 9.9 ng/mg in C2C12 cell extract, 17.3 ng/mg in mouse skeletal muscle extract, 26.7 ng/mg in mouse heart, and 36.7 ng/mg in rat heart.



Interpolated concentrations of PDK4 in various species heart tissue extracts.

Other species reactivity was determined by measuring a 300 µg/mL extract load of various species heart tissue extract, and interpolating the protein concentrations from the mouse standard curve.

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