abcam

Product datasheet

Human PDH E1 alpha protein ELISA Kit (PDHA1) ab181415

SimpleStep ELISA®

1 References 3 Images

Overview

Product name Human PDH E1 alpha protein ELISA Kit (PDHA1)

Detection method Colorimetric

Precision

Intra-assay

Sample	n	Mean	SD	CV%
HeLa extract	2			7.7%

Inter-assay

Sample	n	Mean	SD	CV%
HeLa extract	5			3.8%

Sample type Tissue Extracts, Cell Lysate

Assay type Sandwich (quantitative)

Sensitivity 12 ng/ml

Range 15.62 ng/ml - 1000 ng/ml

Recovery 135 %

Sample specific recovery

Sample type	Average %	Range
Serum	144	129% - 152%
Cell culture media	135	134% - 137%
Fetal Bovine Serum	135	129% - 130%

Assay time 1h 30m

Assay duration One step assay

Species reactivity

Reacts with: Human

Product overview

Human PDH E1 alpha ELISA kit (ab181415) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of PDH E1 alpha protein in human cell and tissue extracts. It uses our proprietary SimpleStep ELISA® technology. Quantitate human PDH E1 alpha with 12 ng/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 SimpeStep ELISA® kits.

Notes

The pyruvate dehydrogenase complex performs the decarboxylation of pyruvate into acetyl CoA, a critical function in metabolism, linking glycolysis and oxidative phosphorylation in mitochondria. The enzyme is composed of multiple copies of three enzymes: pyruvate dehydrogenase (E1), dihydrolipoamide transacetylase (E2) and dihydrolipoamide dehydrogenase (E3). The E1 enzyme is a tetramer of two alpha (PDHA1) and two beta (PDHB) subunits and is present in 30 copies in the PDH complex.

The activity of PDH is regulated by reversible phosphorylation of three serine residues on the PDHA1 subunit at phospho S232, phospho S293, and phospho S300. ELISA assays to measure total PDHA1, phospho 232, phospho S293, and phospho S300 are available from Abcam (ab115342, ab115343, ab115344, ab115345). The phosphorylation of these sites is catalyzed by PDH kinases (PDK). There are four known PDK isoforms, distributed differently in tissues. Their expressions are regulated differently by factors such as starvation, hypoxia and utilization of glucose and fatty acids in various tissues. Dephosporylation, to restore the activity of PDH, is catalyzed by PDH phosphatases (PDP). There are two known isoforms of PDP; PDP1 is present in high levels in skeletal muscle and PDP2 in liver and adipocytes. Functional PDH kinases and phosphatases are available from Abcam (ab110359, ab110354, ab110355, ab110356).

Platform

Microplate

Properties

Storage instructions

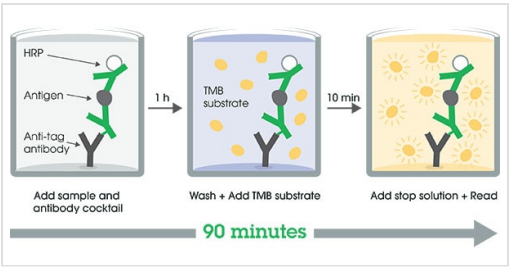
Store at +4°C. Please refer to protocols.

Components	1 x 96 tests
10X Human PDH E1a Capture Antibody	1 x 600µl
10X Human PDH E1a Detector Antibody	1 x 600µl

Components	1 x 96 tests
2X Cell Extraction Buffer LM	1 x 10ml
2X Cell Extraction Buffer LM	1 x 10ml
4X Antibody Diluent EB	1 x 6ml
Human PDH E1 alpha Lyophilized Recombinant Protein	2 vials
Plate Seals	1 unit
Sample Diluent NS (ab193972)	1 x 12ml
SimpleStep Pre-Coated 96-Well Microplate (ab206978)	1 unit
Stop Solution	1 x 12ml
TMB Development Solution	1 x 12ml

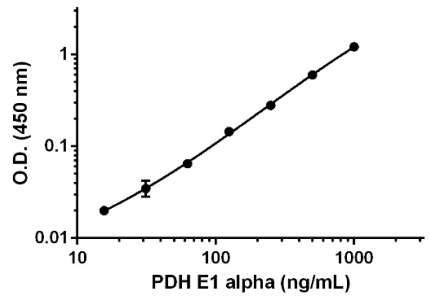
Function	The pyruvate dehydrogenase complex catalyzes the overall conversion of pyruvate to acetyl-CoA and CO(2). It contains multiple copies of three enzymatic components: pyruvate dehydrogenase (E1), dihydrolipoamide acetyltransferase (E2) and lipoamide dehydrogenase (E3).
Tissue specificity	Ubiquitous.
Involvement in disease	Defects in PDHA1 are a cause of pyruvate dehydrogenase E1-alpha deficiency (PDHAD) [MIM:312170]. An enzymatic defect causing primary lactic acidosis in children. It is associated with a broad clinical spectrum ranging from fatal lactic acidosis in the newborn to chronic neurologic dysfunction with structural abnormalities in the central nervous system without systemic acidosis. Defects in PDHA1 are the cause of X-linked Leigh syndrome (X-LS) [MIM:308930]. X-LS is an early-onset progressive neurodegenerative disorder with a characteristic neuropathology consisting of focal, bilateral lesions in one or more areas of the central nervous system, including the brainstem, thalamus, basal ganglia, cerebellum, and spinal cord. The lesions are areas of demyelination, gliosis, necrosis, spongiosis, or capillary proliferation. Clinical symptoms depend on which areas of the central nervous system are involved. The most common underlying cause is a defect in oxidative phosphorylation. LS may be a feature of a deficiency of any of the mitochondrial respiratory chain complexes.
Post-translational modifications	Phosphorylation at Ser-293 by PDK family kinases blocks the access to active site, and inactivates the enzyme.
Cellular localization	Mitochondrion matrix.

Images



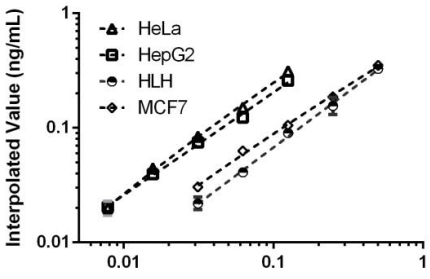
SimpleStep ELISA technology allows the formation of the antibody-antigen complex in one single step, reducing assay time to 90 minutes. Add samples or standards and antibody mix to wells all at once, incubate, wash, and add your final substrate. See protocol for a detailed step-by-step guide.

Other - Human PDH E1 alpha protein ELISA Kit (PDHA1) (ab181415)



Example of PDH E1 alpha (PDHA1) standard curve. Background-subtracted data values (mean +/- SD) are graphed.

Figure 1



Titration of HeLa, HepG2, Human Liver Homogenate (HLH) and MCF7 cell extract within the working range of the assay in 1X Cell Extraction Buffer LM. Background subtracted data from duplicate measurements are plotted.

Figure 2

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