abcam

Product datasheet

Human HIF-2-alpha ELISA Kit ab227898

Recombinant SimpleStep ELISA®

3 References 5 Images

Overview

Product name Human HIF-2-alpha ELISA Kit

Detection method Colorimetric

Precision

Intra-assay

Sample	n	Mean	SD	CV%
HeLa extract	5			2.6%

Inter-assay

Sample	n	Mean	SD	CV%
HeLa extract	3			4%

Sample type Cell culture extracts

Assay type Sandwich (quantitative)

Sensitivity 19 pg/ml

Range 31.3 pg/ml - 2000 pg/ml

Recovery

Sample specific recovery

Sample type	Average %	Range
Cell culture extracts	114	112% - 116%

Assay time 1h 30m

Assay duration One step assay

Species reactivity **Reacts with:** Human

Product overview Human HIF-2-alpha ELISA Kit (ab227898) is a single-wash 90 min sandwich ELISA designed for the quantitative measurement of HIF-2-alpha protein in cell culture extracts. It uses our proprietary SimpleStep ELISA® technology. Quantitate Human HIF-2-alpha with 19 pg/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

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.

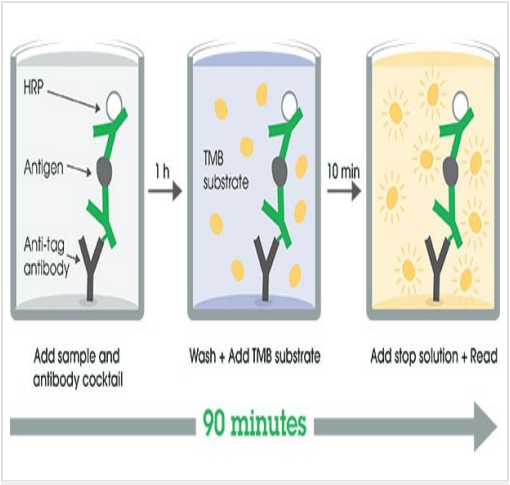
Components	1 x 96 tests
10X Human HIF-2-alpha Capture Antibody	1 x 600µl
10X Human HIF-2-alpha Detector Antibody	1 x 600µl
10X Wash Buffer PT (ab206977)	1 x 20ml
50X Cell Extraction Enhancer Solution (ab193971)	1 x 1ml
5X Cell Extraction Buffer PTR (ab193970)	1 x 10ml
Antibody Diluent CPR	1 x 6ml
Human HIF-2-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

Transcription factor involved in the induction of oxygen regulated genes. Binds to core DNA sequence 5'-[AG]CGTG-3' within the hypoxia response element (HRE) of target gene promoters.

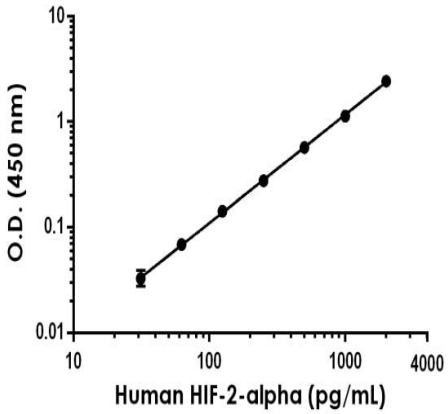
	Regulates the vascular endothelial growth factor (VEGF) expression and seems to be implicated in the development of blood vessels and the tubular system of lung. May also play a role in the formation of the endothelium that gives rise to the blood brain barrier. Potent activator of the Tie-2 tyrosine kinase expression. Activation seems to require recruitment of transcriptional coactivators such as CREBPB and probably EP300. Interaction with redox regulatory protein APEX seems to activate CTAD.
Tissue specificity	Expressed in most tissues, with highest levels in placenta, lung and heart. Selectively expressed in endothelial cells.
Involvement in disease	Defects in EPAS1 are the cause of erythrocytosis familial type 4 (ECYT4) [MIM:611783]. ECYT4 is an autosomal dominant disorder characterized by increased serum red blood cell mass, elevated hemoglobin concentration and hematocrit, and normal platelet and leukocyte counts.
Sequence similarities	Contains 1 basic helix-loop-helix (bHLH) domain. Contains 1 PAC (PAS-associated C-terminal) domain. Contains 2 PAS (PER-ARNT-SIM) domains.
Post-translational modifications	In normoxia, is probably hydroxylated on Pro-405 and Pro-531 by EGLN1/PHD1, EGLN2/PHD2 and/or EGLN3/PHD3. The hydroxylated prolines promote interaction with VHL, initiating rapid ubiquitination and subsequent proteasomal degradation. Under hypoxia, proline hydroxylation is impaired and ubiquitination is attenuated, resulting in stabilization. In normoxia, is hydroxylated on Asn-847 by HIF1AN thus probably abrogating interaction with CREBBP and EP300 and preventing transcriptional activation. Phosphorylated on multiple sites in the CTAD. The iron and 2-oxoglutarate dependent 3-hydroxylation of asparagine is (S) stereospecific within HIF CTAD domains.
Cellular localization	Nucleus.

Images



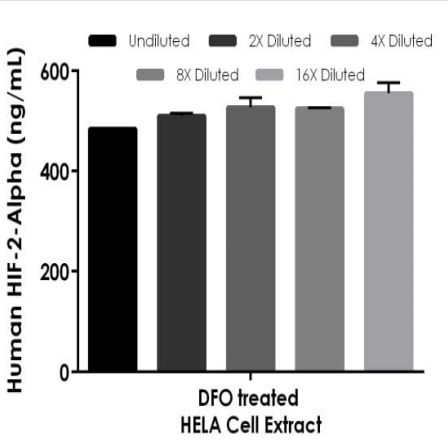
Other - Human HIF-2-alpha ELISA Kit (ab227898)

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.



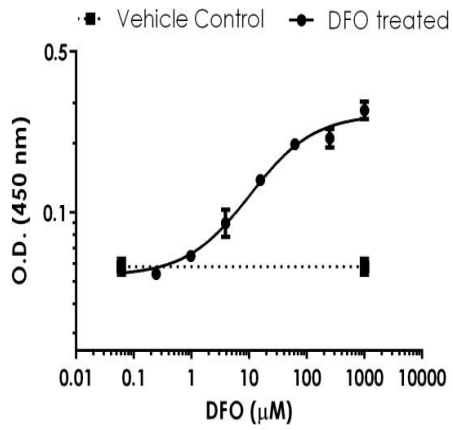
Example of Human HIF-2-alpha standard curve in 1X Cell Extraction Buffer PTR

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



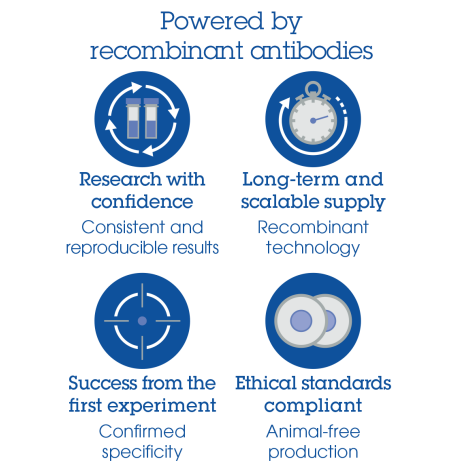
Interpolated concentrations of native HIF-2-alpha in DFO treated Human HeL1 cell extract, sample based on a 250 µg/mL extract load

HELA cells were untreated or treated with 250 µM DFO for 24 hrs, then collected and extracted according to sample preparation protocol (see section 11.1). The concentrations of HIF-2-alpha were measured in duplicate and interpolated from the HIF-2-alpha standard curve and corrected for sample dilution. The interpolated dilution factor corrected values are plotted (mean +/- SD, n=2). The mean HIF-2-alpha concentration was determined to be 520 pg/mL in DFO treated HELA cell extract and undetectable in untreated HELA cell extract.



Dose-dependent induction of HIF-2-alpha in HELA cells by deferoxamine (DFO)

HeLa cells were cultured in 96-well tissue culture plates and were either untreated (Vehicle Control) or exposed to varying doses of DFO for 24 hours (DFO treated). Cells were extracted directly in the culture plate by overlaying culture media with Cell Extraction Buffer PTR with Enhancer such that the final concentration was 1X Cell Extraction Buffer PTR. Extracts were applied to the HIF-2-alpha SimpleStep ELISA plate. Raw data and standard deviation is plotted from quadruplicate measurements.



Sandwich ELISA - Human HIF-2-alpha ELISA Kit (ab227898)

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

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet

- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours

- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.com/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors