


HIF-1 alpha Transcription Factor Assay Kit ab133104

21 References 画像数 4

製品の概要

製品名	HIF-1 alpha Transcription Factor Assay Kit
検出方法	Colorimetric
サンプルの種類	Cell culture extracts, Cell Lysate
アッセイタイプ	Semi-quantitative
種交差性	交差種: Mouse, Rat, Human 交差が予測される動物種: Mammals 

製品の概要	<p>HIF-1 alpha Transcription Factor Assay (ab133104) is a non-radioactive, sensitive method for detecting specific transcription factor DNA binding activity in nuclear extracts and whole cell lysate.</p> <p>A 96-well enzyme-linked immunosorbent assay (ELISA) replaces the cumbersome radioactive electrophoretic mobility shift assay (EMSA). A specific double stranded DNA (dsDNA) sequence containing the HIF-1 alpha response element (5'-ACGTG-3') is immobilized to the wells of a 96-well plate. HIF-1 alpha contained in a nuclear extract, binds specifically to the HIF-1 alpha response element. The HIF transcription factor complex is detected by addition of a specific primary antibody directed against HIF-1 alpha. A secondary antibody conjugated to HRP is added to provide a sensitive colorimetric readout at 450 nm.</p>
試験プラットフォーム	Microplate reader

製品の特性

保存方法	Please refer to protocols.
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内容	96 tests
96-Well Plate Cover	1 unit
Polysorbate 20	1 vial
Transcription Factor Antibody Binding Buffer (10X)	1 x 3ml
Transcription Factor Binding Assay Buffer (4X)	1 x 3ml

内容	96 tests
Transcription Factor Developing Solution	1 x 12ml
Transcription Factor Goat Anti-Rabbit HRP Conjugate	1 x 100µl
Transcription Factor HIF-1 alpha 96-Well Strip Plate	1 unit
Transcription Factor HIF-1 alpha Competitor dsDNA	1 vial
Transcription Factor HIF-1 alpha Positive Control	1 vial
Transcription Factor HIF-1 alpha Primary Antibody	1 vial
Transcription Factor Reagent A	1 x 120µl
Transcription Factor Stop Solution	1 x 12ml
Wash Buffer Concentrate (400X)	1 x 5ml

<b>機能</b>	Functions as a master transcriptional regulator of the adaptive response to hypoxia. Under hypoxic conditions activates the transcription of over 40 genes, including, erythropoietin, glucose transporters, glycolytic enzymes, vascular endothelial growth factor, and other genes whose protein products increase oxygen delivery or facilitate metabolic adaptation to hypoxia. Plays an essential role in embryonic vascularization, tumor angiogenesis and pathophysiology of ischemic disease. Binds to core DNA sequence 5'-[AG]CGTG-3' within the hypoxia response element (HRE) of target gene promoters. Activation requires recruitment of transcriptional coactivators such as CREBPB and EP300. Activity is enhanced by interaction with both, NCOA1 or NCOA2. Interaction with redox regulatory protein APEX seems to activate CTAD and potentiates activation by NCOA1 and CREBBP.
<b>組織特異性</b>	Expressed in most tissues with highest levels in kidney and heart. Overexpressed in the majority of common human cancers and their metastases, due to the presence of intratumoral hypoxia and as a result of mutations in genes encoding oncoproteins and tumor suppressors.
<b>配列類似性</b>	Contains 1 basic helix-loop-helix (bHLH) domain. Contains 1 PAC (PAS-associated C-terminal) domain. Contains 2 PAS (PER-ARNT-SIM) domains.
<b>ドメイン</b>	Contains two independent C-terminal transactivation domains, NTAD and CTAD, which function synergistically. Their transcriptional activity is repressed by an intervening inhibitory domain (ID).
<b>翻訳後修飾</b>	In normoxia, is hydroxylated on Pro-402 and Pro-564 in the oxygen-dependent degradation domain (ODD) by EGLN1/PHD1 and EGLN2/PHD2. EGLN3/PHD3 has also been shown to hydroxylate Pro-564. The hydroxylated prolines promote interaction with VHL, initiating rapid ubiquitination and subsequent proteasomal degradation. Deubiquitinated by USP20. Under hypoxia, proline hydroxylation is impaired and ubiquitination is attenuated, resulting in stabilization. In normoxia, is hydroxylated on Asn-803 by HIF1AN, thus abrogating interaction with CREBBP and EP300 and preventing transcriptional activation. This hydroxylation is inhibited by the Cu/Zn-chelator, Clioquinol. S-nitrosylation of Cys-800 may be responsible for increased recruitment of p300 coactivator necessary for transcriptional activity of HIF-1 complex. Requires phosphorylation for DNA-binding. Sumoylated; by SUMO1 under hypoxia. Sumoylation is enhanced through interaction with

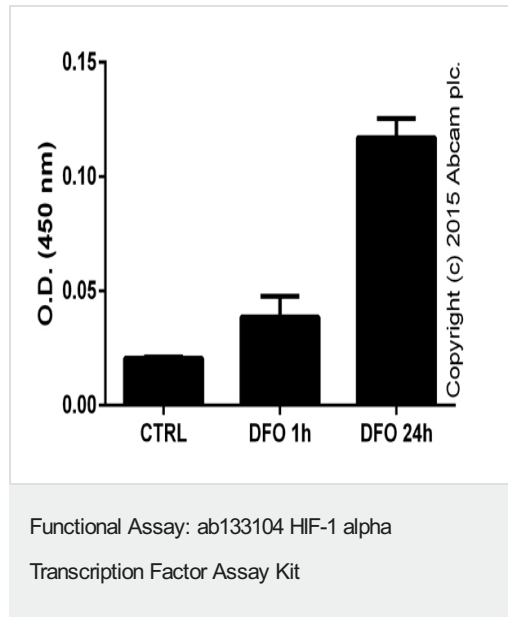
RWDD3. Desumoylation by SENP1 leads to increased HIF1A stability and transcriptional activity. Ubiquitinated; in normoxia, following hydroxylation and interaction with VHL. Lys-532 appears to be the principal site of ubiquitination. Clioquinol, the Cu/Zn-chelator, inhibits ubiquitination through preventing hydroxylation at Asn-803.

The iron and 2-oxoglutarate dependent 3-hydroxylation of asparagine is (S) stereospecific within HIF CTAD domains.

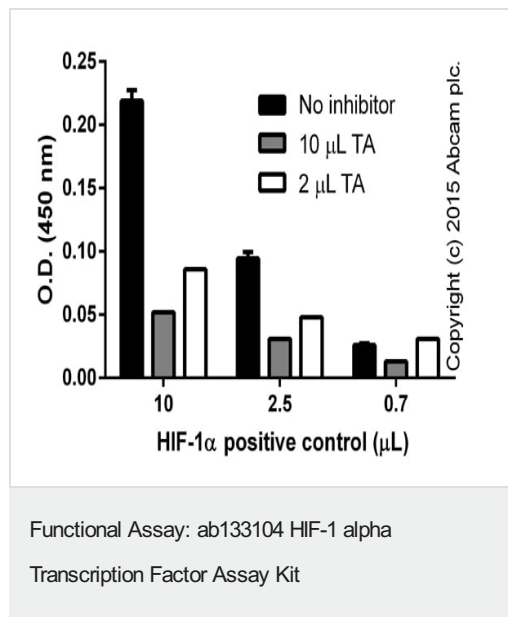
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Cytoplasm. Nucleus. Cytoplasmic in normoxia, nuclear translocation in response to hypoxia. Colocalizes with SUMO1 in the nucleus, under hypoxia.

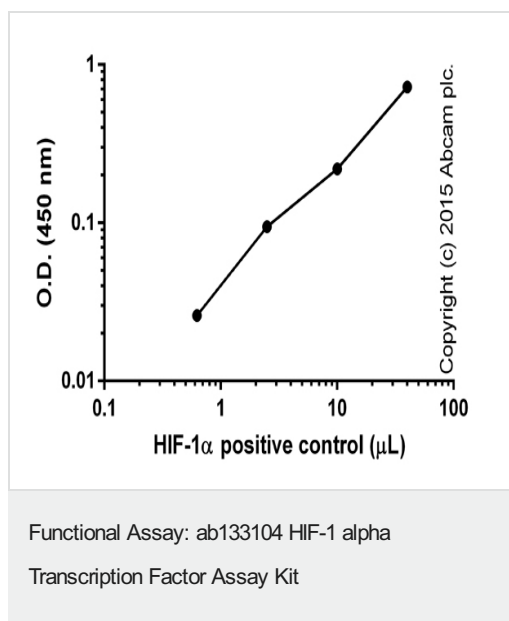
## 画像



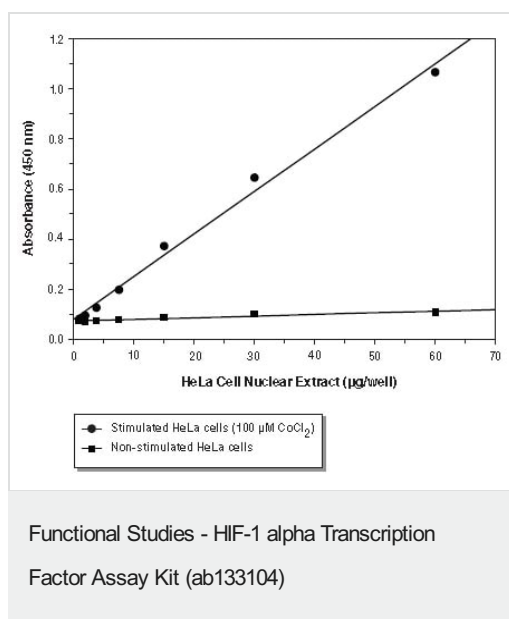
HEK293 cells were treated with 1 mM deferoxamine mesylate (DFO) for 1 or 24 hours. 40 microliters of nuclear lysates ([ab113474](#); corresponding to 4e6 cells) were tested in duplicates (+/- SD).



Titration of positive control with different volumes of inhibitor (TA), background signal subtracted (duplicates; +/- SD).



Titration of positive control, background signal subtracted (duplicates; +/- SD).



Assay of nuclear extract from stimulated HeLa cells (100 μM CoCl<sub>2</sub>) and non-stimulated HeLa cells.

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