

# Anti-HIF-2-alpha antibody [ep190b] ab8365



★★★★☆ 2 Abreviews | 28 References | 4 Images

## Overview

Product name	Anti-HIF-2-alpha antibody [ep190b]
Description	Mouse monoclonal [ep190b] to HIF-2-alpha
Specificity	This antibody is specific for HIF-2-alpha and does not cross-react with HIF-1-alpha.
Tested applications	Flow Cyt, ELISA, IHC-Fr, IHC-P, WB
Species reactivity	<b>Reacts with:</b> Rat, Human
Immunogen	Fusion protein (proprietary-tag) corresponding to Human HIF-2-alpha aa 535-631.
Positive control	Hypoxic A549 lysate.

## Properties

Form	Liquid
Storage instructions	Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.1% Sodium azide
Purity	Protein G purified
Clonality	Monoclonal
Clone number	ep190b
Myeloma	NS1
Isotype	IgG1

## Applications

Our [Abpromise guarantee](#) covers the use of **ab8365** 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
<a href="#">Flow Cyt</a>		Use 2µg for 10 <sup>6</sup> cells. Ab81216-Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.
<a href="#">ELISA</a>		Use at an assay dependent concentration.
<a href="#">IHC-Fr</a>		Use at an assay dependent concentration.
<a href="#">IHC-P</a>		1/1000 - 1/3000. Perform heat mediated antigen retrieval with Tris/EDTAbuffer pH 9.0 before commencing with IHC staining protocol.
<a href="#">WB</a>	★★★★☆	1/100 - 1/500. Detects a band of approximately 118 kDa (predicted molecular weight: 100 kDa).

## Target

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.

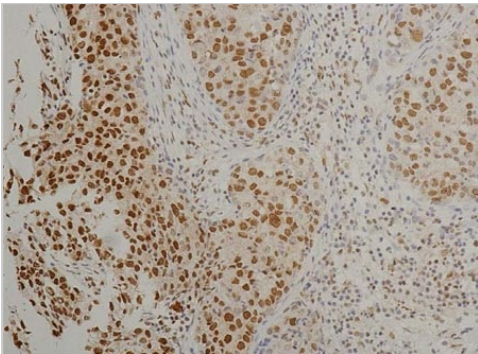
Post-translational modifications

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.

Anti-HIF-2-alpha antibody [ep190b] images

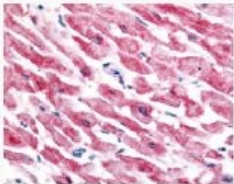


Immunohistochemical analysis of Human breast cancer tissue, staining HIF-2-alpha with ab8365.

Antigen retrieval was performed using EDTA buffer in a pressure cooker, before being blocked with normal serum. Samples were incubated with primary antibody (1/2000) overnight at 4°C. Staining was detected using DAB.

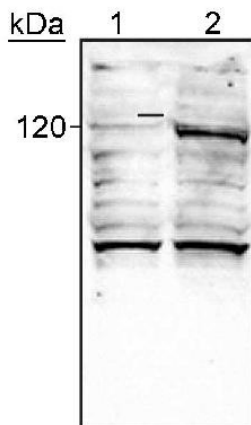
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF-2-alpha [ep190b] antibody (ab8365)

Image from Xiang L. et al., Diagn Pathol. 2012 Mar 27;7:32. Fig 5.; doi:10.1186/1746-1596-7-32; 27 March 2012, Diagnostic Pathology 2012, 7:32



ab8365 at 1/300 dilution, staining HIF-2-alpha in cardiomyocytes of Human heart tissue section by Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections).

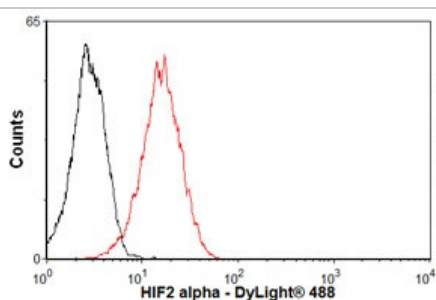
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF-2-alpha [ep190b] antibody (ab8365)



Western blot - Anti-HIF-2-alpha [ep190b] antibody (ab8365)

**Predicted band size :** 100 kDa

HIF-2-alpha detected in hypoxic Human lysate using ab8365. Lane 1: normoxic A549 lysate control, lane 2: hypoxic A549 lysate.



Flow Cytometry - Anti-HIF-2-alpha [ep190b] antibody (ab8365)

Overlay histogram showing HepG2 cells stained with ab8365 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab8365, 2µg/1x10<sup>6</sup> cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10<sup>6</sup> cells) used under the same conditions. Acquisition of >5,000 events was performed.

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