abcam

Product datasheet

Anti-HIF-2-alpha antibody ab109616

敲除验证

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概述

产品名称	Anti-HIF-2-alpha**抗体**
描述	**兔多克隆抗体**to HIF-2-alpha
宿主	Rabbit
特异性	From Jan 2024, QC testing of replenishment batches of this polyclonal changed. All tested and expected application and reactive species combinations are still covered by our Abcam product promise. However, we no longer test all applications. For more information on a specific batch, please contact our Scientific Support who will be happy to help.
经测试应用	**适用于:** WB, IHC-P, ICC/IF
种属反应性	**与反应:** Mouse, Rat, Human **预测可用于:** Chimpanzee, Macaque monkey, Chinese hamster, Orangutan
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
阳性对照	WB: Wild-type A549 + DFO (1mM,24 hours) cell lysate. IHC: Human colon tissue. ICC/IF: HeLa cells (untreated and treated with Deferoxamine).
常规说明	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

性能

形式	Liquid
存放说明	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -80°C. Avoid freeze / thaw cycle.
存储溶液	pH: 7.40 Preservative: 0.02% Sodium azide Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising agent. If you would like information about the formulation of a specific lot, please contact our scientific support team who will be happy to help.

纯度	Immunogen affinity purified
克隆	**多克隆**
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab109616于以下的经测试应用**

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★☆☆ (2)	Use a concentration of 1 µg/ml. Detects a band of approximately 100 kDa (predicted molecular weight: 96 kDa).
IHC-P		Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use a concentration of 10 µg/ml.

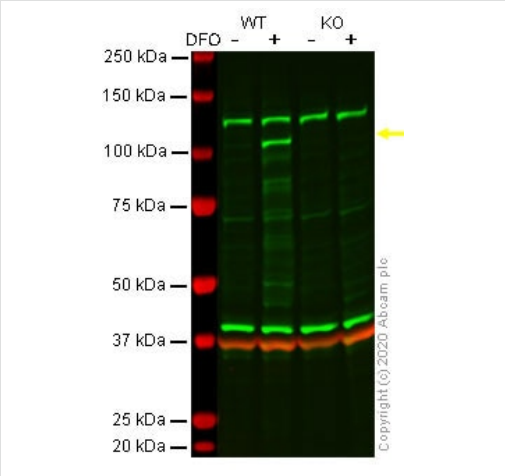
靶标

功能	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.
组织特异性	Expressed in most tissues, with highest levels in placenta, lung and heart. Selectively expressed in endothelial cells.
疾病相关	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.
序列相似性	Contains 1 basic helix-loop-helix (bHLH) domain. Contains 1 PAC (PAS-associated C-terminal) domain. Contains 2 PAS (PER-ARNT-SIM) domains.
翻译后修饰	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.

细胞定位 Nucleus.

图片



Western blot - Anti-HIF-2-alpha antibody (ab109616)

All lanes : Anti-HIF-2-alpha antibody (ab109616) at 1 µg/ml

Lane 1 : Wild-type A549 untreated cell lysate
Lane 2 : Wild-type A549 + DFO (1mM, 24 hours) cell lysate
Lane 3 : EPAS knockout A549 untreated cell lysate
Lane 4 : EPAS knockout A549 + DFO (1mM, 24 hours) cell lysate

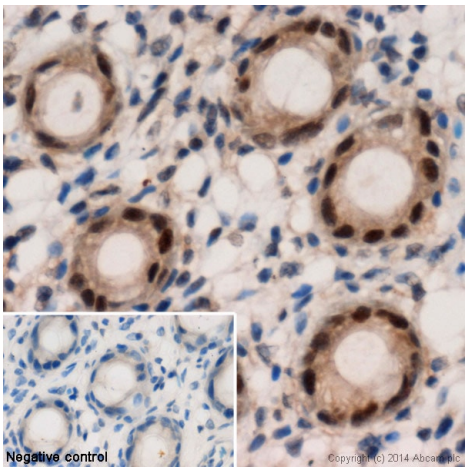
Lysates/proteins at 40 µg per lane.

Performed under reducing conditions.

Predicted band size: 96 kDa
Observed band size: 100 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab109616 observed at 100 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

ab109616 was shown to react with HIF-2-alpha in A549 wild-type cells in western blot with loss of signal observed in EPAS1 knockout sample. A549 wild-type and EPAS1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab109616 and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at 1 µg/ml and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

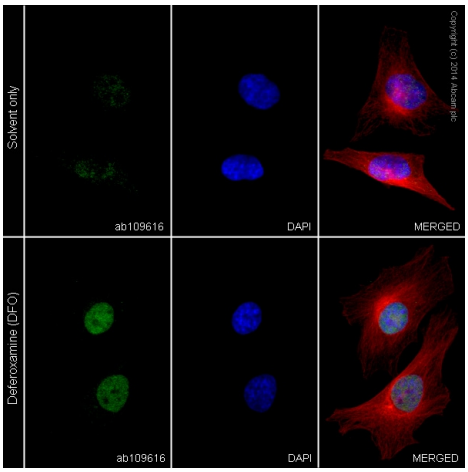


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF-2-alpha antibody (ab109616)

IHC image of HIF-2alpha staining in a section of formalin-fixed paraffin-embedded normal human colon*, performed on a Leica BOND. The section was pre-treated using heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 for 20mins. The section was then incubated with ab109616 at 1µg/ml, for 15 mins at room temperature. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset negative control image is taken from an identical assay without primary antibody.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Immunocytochemistry/ Immunofluorescence - Anti-HIF-2-alpha antibody (ab109616)

ab109616 staining Hif-2-alpha in HeLa cells. The cells were incubated with 1mM Deferoxamine for 24 hours (Treated) or solvent-only for control purposes (Solvent only). Cells were fixed with 4% formaldehyde (10min) and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated with ab109616 at 10µg/ml and **ab195889** at 1µg/ml overnight at +4°C. Nuclear DNA was labelled in blue with DAPI.

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