abcam

ab16066

Anti-HIF-1 alpha antibody [mgc3]

Anti-HIF-1 alpha antibody [mgc3] (ab16066) is a mouse monoclonal antibody detecting HIF-1 alpha in **Western Blot, Flow Cytometry, IHC-P**. Suitable for **Human**.

- Over 140 publications
- Trusted since 2004

Key facts

Isotype	IgG1
Host species	Mouse
Storage buffer	pH: 7.4 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	Recombinant Fragment Protein within Human HIF1A aa 500-850. The exact immunogen used to generate this antibody is proprietary information. Database link Q16665
Clone number	mgc3
Purification technique	Affinity purification Protein G
Specificity	This antibody does not cross-react with ARNT or the related HIF-2-alpha.
Concentration	1 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/100
Notes	Perform heat-mediated antigen retrieval before commencing with IHC staining protocol.

Predicted

Species	Mouse, Cow, Pig, Primates
Dilution info	-
Notes	-

Flow Cyt

Tested

Species	Human
Dilution info	2 µg for 10^6 Cells
Notes	ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Predicted

Species	Mouse, Cow, Pig, Primates
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/2000
Notes	-

Predicted

Species	Mouse, Cow, Pig, Primates
Dilution info	-
Notes	-

Target data

See full target information HIF1A

Function	Functions as a master transcriptional regulator of the adaptive response to hypoxia (PubMed:11292861, PubMed:11566883, PubMed:15465032, PubMed:16973622, PubMed:17610843, PubMed:18658046, PubMed:20624928, PubMed:22009797, PubMed:30125331, PubMed:9887100). Under hypoxic conditions, activates the transcription of over 40 genes, including erythropoietin, glucose transporters, glycolytic enzymes, vascular endothelial growth factor, HILPDA, and other genes whose protein products increase oxygen delivery or facilitate metabolic adaptation to hypoxia (PubMed:11292861, PubMed:11566883, PubMed:15465032, PubMed:16973622, PubMed:17610843, PubMed:20624928, PubMed:22009797, PubMed:30125331, PubMed:9887100). Plays an essential role in embryonic vascularization, tumor angiogenesis and pathophysiology of ischemic disease (PubMed:22009797). Heterodimerizes with ARNT; heterodimer binds to core DNA sequence 5'-TACGTG-3' within the hypoxia response element (HRE) of target gene promoters (By similarity). Activation requires recruitment of transcriptional coactivators such as CREBBP and EP300 (PubMed:16543236, PubMed:9887100). Activity is enhanced by interaction with NCOA1 and/or NCOA2 (PubMed:10594042). Interaction with redox regulatory protein APEX1 seems to activate CTAD and potentiates activation by NCOA1 and CREBBP (PubMed:10202154, PubMed:10594042). Involved in the axonal distribution and transport of mitochondria in neurons during hypoxia (PubMed:19528298). (Microbial infection) Upon infection by human coronavirus SARS-CoV-2, is required for induction of glycolysis in monocytes and the consequent pro-inflammatory state (PubMed:32697943). In monocytes, induces expression of ACE2 and cytokines such as IL1B, TNF, IL6, and interferons (PubMed:32697943). Promotes human coronavirus SARS-CoV-2 replication and monocyte inflammatory response (PubMed:32697943).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C

Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-HIF-1 alpha antibody [mgc3] (ab16066) is a mouse monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Flow Cyt), Immunohistochemistry (IHC-P) in Human samples.

What is the molecular weight of HIF-1 alpha?
Anti-HIF-1 alpha [mgc3] (ab16066) specifically detects a band for HIF-1 alpha (UniProt: Q16665) at a molecular weight of 92kDa.

Trusted by the scientific community
Anti-HIF-1 alpha [mgc3] (ab16066) was first used in a scientific publication in 2004 and has been cited over 140 times in peer-reviewed journals.

Reviewed by scientists
Anti-HIF-1 alpha [mgc3] (ab16066) has over 10 independent reviews from customers.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	HIF-1 alpha also known as hypoxia-inducible factor 1-alpha is a transcription factor critical in cellular response to low oxygen levels. Its molecular weight usually ranges from 93 to 120 kDa. You can find HIF-1 alpha expressed in tissues throughout the body but its expression significantly increases under hypoxic conditions. Researchers often use the HIF-1a ELISA to measure its expression levels. HIF-1 alpha forms a complex with other proteins to perform its functions effectively.
Biological function summary	HIF-1 alpha regulates gene expression in response to hypoxic conditions in cells. It forms a complex with HIF-1 beta to activate transcription of various genes involved in energy metabolism angiogenesis and erythropoiesis. HIF-1 alpha enables cells to adapt to reduced oxygen availability allowing for cellular survival and function under stress. It plays an important role in promoting the expression of genes like VEGF and EPO which are important for vascular and red blood cell development respectively.
Pathways	HIF-1 alpha plays an integral role in the hypoxia signaling pathway and the glycolytic pathway. In the hypoxia signaling pathway HIF-1 alpha partners with VHL (Von Hippel-Lindau) protein that regulates its degradation under normal oxygen conditions. When oxygen levels drop HIF-1 alpha avoids degradation stabilizes and translocates into the nucleus to initiate transcription of hypoxia-responsive genes. The glycolytic pathway involvement highlights its function in adapting energy production under hypoxic conditions through collaboration with enzymes and transporters associated with glycolysis.

Associated diseases and disorders	HIF-1 alpha has been implicated in cancer and ischemic diseases. Its role in promoting angiogenesis and metabolic adaptation makes it a contributor to tumor growth and survival collaborating with oncogenes such as c-Myc. In ischemic diseases like stroke or myocardial infarction HIF-1 alpha's ability to induce protective responses can mitigate tissue damage through regulation of survival pathways. Understanding these interactions helps in the development of therapeutic strategies targeting HIF-1 alpha in disease contexts.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

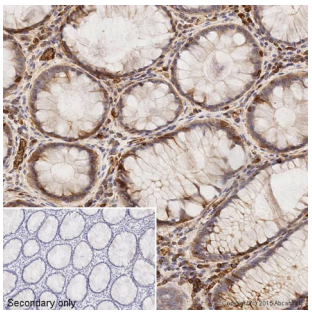
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

5 product images

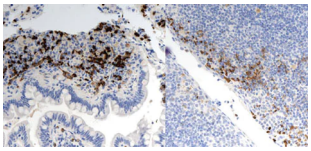


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF-1 alpha antibody [mgc3] (ab16066)

HIF-1 alpha Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-HIF-1 alpha antibody

IHC image of ab16066 staining HIF-1 alpha in human colon formalin fixed paraffin embedded tissue sections*, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with Tris/EDTA buffer (pH9, epitope retrieval solution 2) for 20 mins. The section was then incubated with ab16066, 10µg/ml working concentration, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as

the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. No primary antibody was used in the secondary only control (shown on the inset).
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

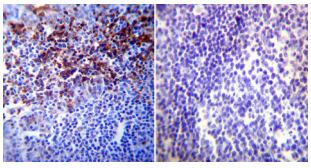


This image is courtesy of an anonymous customer review

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF-1 alpha antibody [mgc3] (ab16066)

HIF-1 alpha Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-HIF-1 alpha antibody

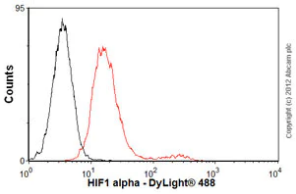
ab16066 staining HIF-1-alpha in Human small intestine (IBD) and tonsil tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with formaldehyde; antigen retrieval was by heat mediation with an EDTA buffer (pH 9.0). Samples were incubated with primary antibody (1/800 in diluent + background reducers) for 20 minutess at 25°C. An undiluted Goat polymer was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF-1 alpha antibody [mgc3] (ab16066)

HIF-1 alpha Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using mouse Anti-HIF-1 alpha antibody

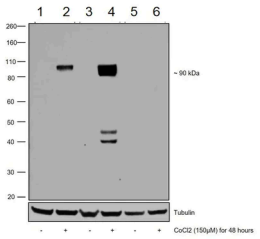
Immunohistochemistry was performed on normal biopsies of deparaffinized Human tonsil tissue. To expose target proteins heat induced antigen retrieval was performed using 10mM sodium citrate (pH6.0) buffer microwaved for 8-15 minutes. Following antigen retrieval tissues were blocked in 3% BSA-PBS for 30 minutes at room temperature. Tissues were then probed at a dilution of 1/20 with ab16066 (left) or without primary antibody (negative control - right) overnight at 4°C in a humidified chamber. Tissues were washed extensively with PBST and endogenous peroxidase activity was quenched with a peroxidase suppressor. Detection was performed using a biotin-conjugated secondary antibody and SA-HRP followed by colorimetric detection using DAB. Tissues were counterstained with hematoxylin and prepped for mounting.



Flow Cytometry - Anti-HIF-1 alpha antibody [mgc3] (ab16066)

HIF-1 alpha Flow Cytometry staining using mouse Anti-HIF-1 alpha antibody

Overlay histogram showing HeLa cells stained with ab16066 (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 (ab16066, 2µg/1x10^6 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/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.



Western blot - Anti-HIF-1 alpha antibody [mgc3] (ab16066)

HIF-1 alpha Western blot staining using mouse Anti-HIF-1 alpha antibody

Antibody specificity was demonstrated by CRISPR-Cas9 mediated knockout of target protein. A loss of signal was observed for target protein in HIF-1 alpha KO cell line compared to control cell lines using ab16066.
Uncharacterized bands were observed in HeLa Cas9 samples at ~40 kDa and 45 kDa.

All lanes:
Western blot - Anti-HIF-1 alpha antibody [mgc3] (ab16066) at 1/2000 dilution

Lane 1:
HeLa wild type (untreated) at 30 µg

Lane 2:
HeLa wild type treated with 150uM CoCl2 for 48 hrs at 30 µg

Lane 3:
HeLa Cas9 at 30 µg

Lane 4:
HeLa Cas9 treated with 150uM CoCl2 for 48 hrs at 30 µg

Lane 5:
HeLa HIF1A KO at 30 µg

Lane 6:
HeLa HIF1A KO treated with 150uM CoCl2 for 48 hrs at 30 µg

Secondary

All lanes:
Goat anti-Mouse IgG (H+L) Superclonal™ Recombinant Secondary Antibody, HRP at 1/4000 dilution

Predicted band size: 92 kDa

Observed band size: 90 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.