

Product datasheet

Anti-HIF1 beta antibody [EPR23106-151] ab270520

KO VALIDATED Recombinant RabMAb

8 Images

Overview

Product name	Anti-HIF1 beta antibody [EPR23106-151]
Description	Rabbit monoclonal [EPR23106-151] to HIF1 beta
Host species	Rabbit
Tested applications	Suitable for: Flow Cyt (Intra), WB, IHC-P Unsuitable for: ChIP, ICC/IF or IP
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: MDA-MB-231, RAW264.7, 293T, C6, HAP1 and HeLa cell lysates. IHC-P: Human colon tissue; Mouse testis tissue; Rat testis tissue. Flow Cyt (intra): HeLa and NIH/3T3 cells.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none">- High batch-to-batch consistency and reproducibility- Improved sensitivity and specificity- Long-term security of supply- Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR23106-151

Isotype

IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab270520 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
Flow Cyt (Intra)		1/600.
WB		1/1000. Detects a band of approximately 87 kDa (predicted molecular weight: 87 kDa).
IHC-P		1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Application notes

Is unsuitable for ChIP, ICC/IF or IP.

Target

Function

Required for activity of the Ah (dioxin) receptor. This protein is required for the ligand-binding subunit to translocate from the cytosol to the nucleus after ligand binding. The complex then initiates transcription of genes involved in the activation of PAH procarcinogens. The heterodimer with HIF1A or EPAS1/HIF2A functions as a transcriptional regulator of the adaptive response to hypoxia.

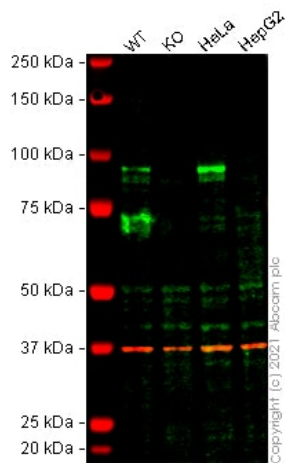
Sequence similarities

Contains 1 basic helix-loop-helix (bHLH) domain.
Contains 1 PAC (PAS-associated C-terminal) domain.
Contains 2 PAS (PER-ARNT-SIM) domains.

Cellular localization

Nucleus.

Images



Western blot - Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

All lanes : Anti-HIF1 beta antibody [EPR23106-151] (ab270520) at 1/1000 dilution

Lane 1 : Wild-type HAP1 cell lysate

Lane 2 : ARNT knockout HAP1 cell lysate

Lane 3 : HeLa cell lysate

Lane 4 : HepG2 cell lysate

Lysates/proteins at 20 µg per lane.

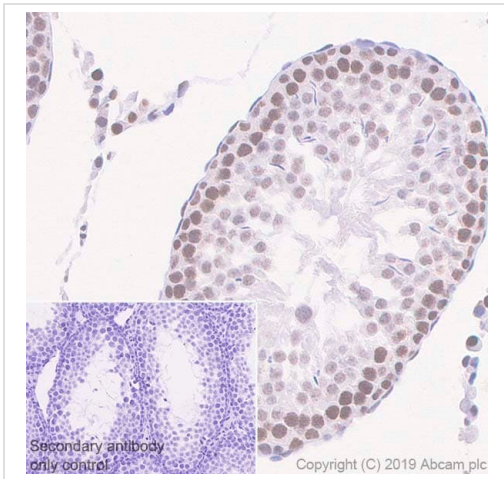
Performed under reducing conditions.

Predicted band size: 87 kDa

Observed band size: 90 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab270520 observed at 90 kDa. Red - loading control **ab8245** (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab270520 was shown to react with HIF1 beta in wild-type HAP1 cells in Western blot with loss of signal observed in ARNT knockout sample. Wild-type HAP1 and ARNT knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 5 % milk in TBS-T (0.1 % Tween[®]) before incubation with ab270520 and **ab8245** (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 dilution 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 h at room temperature before imaging.

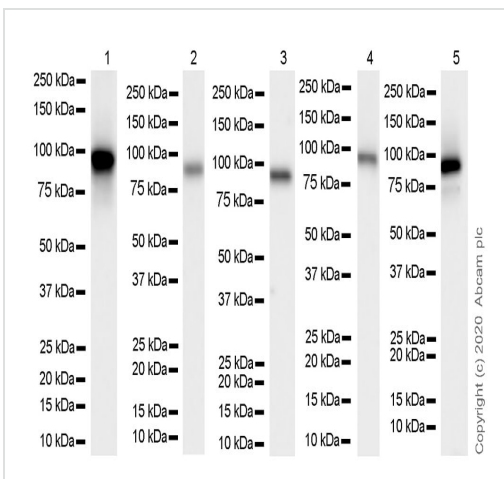


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

Immunohistochemical analysis of paraffin-embedded Rat testis tissue labeling HIF1 beta with ab270520 at 1/500 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining on rat testis is observed. The section was incubated with ab270520 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**).



Western blot - Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

All lanes : Anti-HIF1 beta antibody [EPR23106-151] (ab270520) at 1/1000 dilution

Lane 1 : MDA-MB-231 (human breast adenocarcinoma epithelial cell), whole cell lysate

Lane 2 : RAW264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage), whole cell lysate

Lane 3 : 293T (human embryonic kidney epithelial cell), whole cell lysate

Lane 4 : C6 (rat glial tumor glial cell), whole cell lysate

Lane 5 : HeLa (human cervix adenocarcinoma epithelial cell), whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 87 kDa

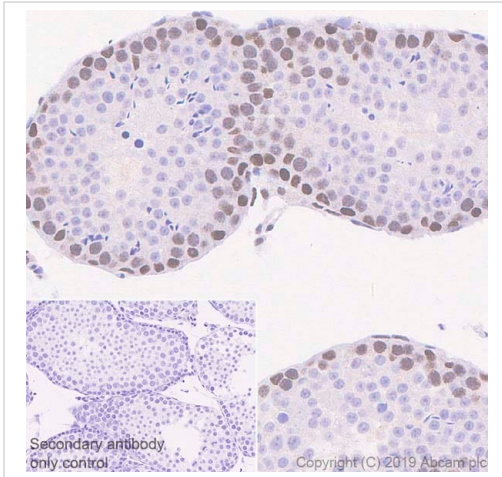
Observed band size: 87 kDa

Exposure time: 26 seconds

Blocking and dilution buffer: 5% NFDm/TBST.

Lysates should be made fresh and used in WB immediately to

minimize protein degradation.

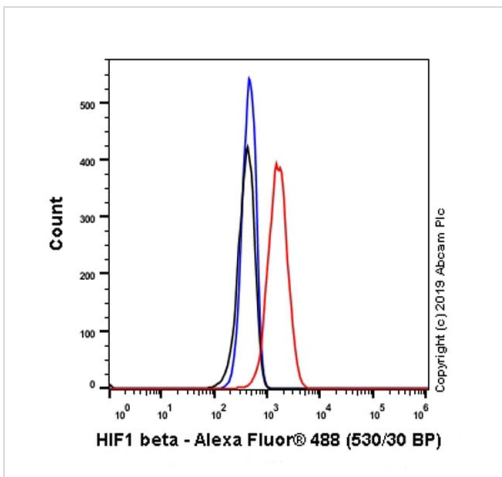


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

Immunohistochemical analysis of paraffin-embedded Mouse testis tissue labeling HIF1 beta with ab270520 at 1/500 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining on mouse testis is observed. The section was incubated with ab270520 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with Hematoxylin.

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.

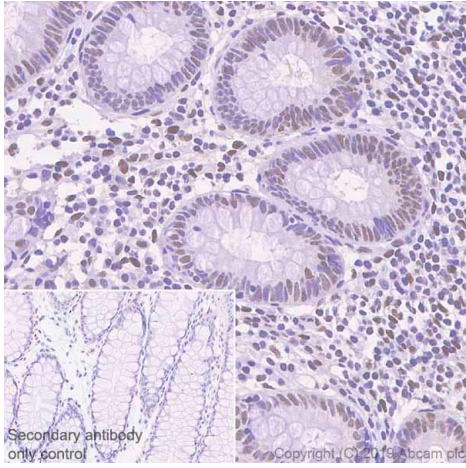
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**).



Flow Cytometry (Intracellular) - Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized HeLa (Human cervix adenocarcinoma epithelial cell) cells labelling HIF1 beta with ab270520 at 1/600 dilution (0.1 µg) (Red) compared with a Rabbit monoclonal IgG (**ab172730**) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).

Goat anti rabbit IgG (Alexa Fluor[®] 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

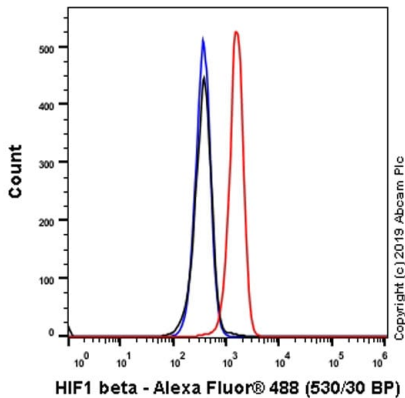


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling HIF1 beta with ab270520 at 1/500 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**). Nuclear staining on human colon is observed. The section was incubated with ab270520 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument. Counterstained with Hematoxylin.

Heat mediated antigen retrieval with Citrate buffer (pH 6.0, epitope retrieval solution 1) for 20 mins.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (**ab209101**).



Flow Cytometry (Intracellular) - Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized NIH/3T3 (Mouse embryonic fibroblast) cells labelling HIF1 beta with ab270520 at 1/600 dilution (0.1 µg) (Red) compared with a Rabbit monoclonal IgG (**ab172730**) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).

A Goat anti rabbit IgG (Alexa Fluor[®] 488, **ab150077**) at 1/2000 dilution was used as the secondary antibody.

Why choose a recombinant antibody?

 Research with confidence Consistent and reproducible results	 Long-term and scalable supply Recombinant technology
 Success from the first experiment Confirmed specificity	 Ethical standards compliant Animal-free production

Anti-HIF1 beta antibody [EPR23106-151] (ab270520)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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