abcam

Product datasheet

Anti-PHD2 / prolyl hydroxylase antibody [EPR3660(B)(2)] ab133630

KO VALIDATED Recombinant RabMAb

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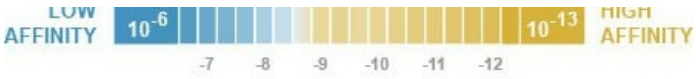
Overview

Product name	Anti-PHD2 / prolyl hydroxylase antibody [EPR3660(B)(2)]
Description	Rabbit monoclonal [EPR3660(B)(2)] to PHD2 / prolyl hydroxylase
Host species	Rabbit
Tested applications	**Suitable for:** WB, IP **Unsuitable for:** ICC/IF or IHC-P
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide within Human PHD2/ prolyl hydroxylase (N terminal). The exact sequence is proprietary.
Positive control	SH-SY5Y treated with cobalt chloride, SH-SY5Y, and Human adrenal gland lysates
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

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.
Dissociation constant (K_D)	$K_D = 1.43 \times 10^{-11}$ M



Learn more about K_D

Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR3660(B)(2)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab133630 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

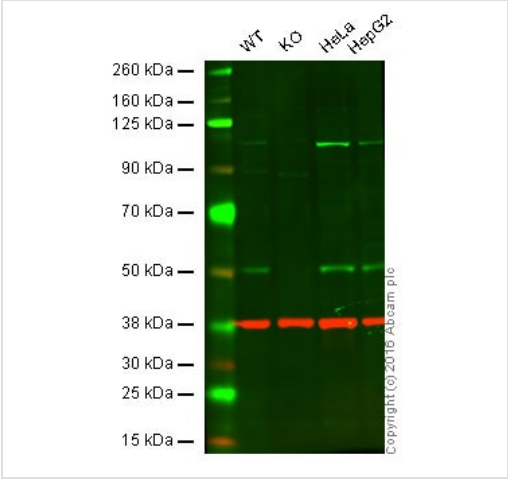
Application	Abreviews	Notes
WB	★★★★☆ (1)	1/1000 - 1/10000. Predicted molecular weight: 46 kDa.
IP		1/10 - 1/100.

Application notes Is unsuitable for ICC/IF or IHC-P.

Target

Function	Catalyzes the post-translational formation of 4-hydroxyproline in hypoxia-inducible factor (HIF) alpha proteins. Hydroxylates HIF-1 alpha at 'Pro-402' and 'Pro-564', and HIF-2 alpha. Functions as a cellular oxygen sensor and, under normoxic conditions, targets HIF through the hydroxylation for proteasomal degradation via the von Hippel-Lindau ubiquitination complex.
Tissue specificity	According to PubMed:11056053, widely expressed with highest levels in skeletal muscle and heart, moderate levels in pancreas, brain (dopaminergic neurons of adult and fetal substantia nigra) and kidney, and lower levels in lung and liver. According to PubMed:12351678 widely expressed with highest levels in brain, kidney and adrenal gland. Expressed in cardiac myocytes, aortic endothelial cells and coronary artery smooth muscle.
Involvement in disease	Defects in EGLN1 are the cause of erythrocytosis familial type 3 (ECYT3) [MIM:609820]. ECYT3 is an autosomal dominant disorder characterized by increased serum red blood cell mass, elevated serum hemoglobin and hematocrit, and normal serum erythropoietin levels.
Sequence similarities	Contains 1 Fe2OG dioxygenase domain. Contains 1 MYND-type zinc finger.

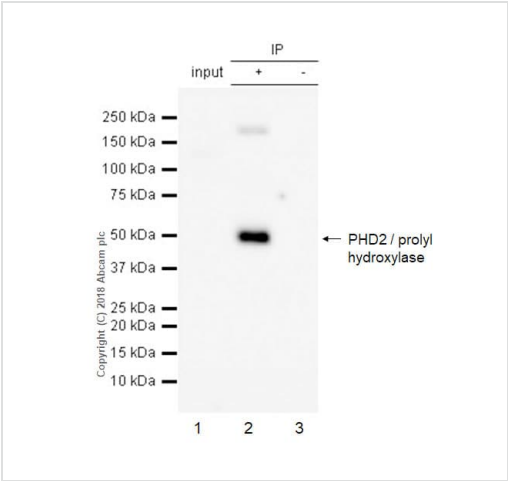
Images



Western blot - Anti-PHD2 / prolyl hydroxylase antibody [EPR3660(B)(2)] (ab133630)

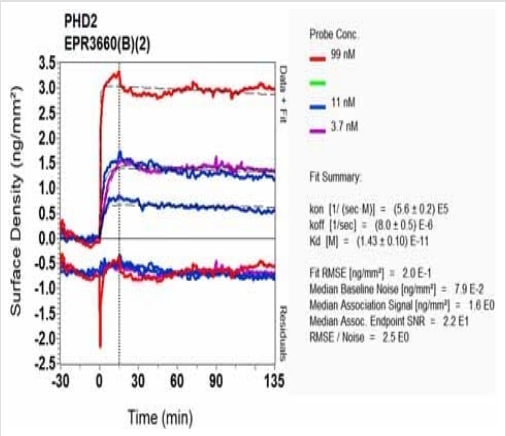
Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: PHD2 / prolyl hydroxylase knockout HAP1 cell lysate (20 µg)
Lane 3: HeLa cell lysate (20 µg)
Lane 4: HepG2 cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab133630 observed at 1X kDa. Red - loading control, **ab8245**, observed at 37 kDa.

Unpurified ab133630 was shown to specifically react with PHD2 / prolyl hydroxylase when PHD2 / prolyl hydroxylase knockout samples were used. Wild-type and PHD2 / prolyl hydroxylase knockout samples were subjected to SDS-PAGE. Unpurified ab133630 and **ab8245** (loading control to GAPDH) were diluted 1/1000 and 1/10000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.



Immunoprecipitation - Anti-PHD2 / prolyl hydroxylase antibody [EPR3660(B)(2)] (ab133630)

ab133630 (purified) at 1:40 dilution (2µg) immunoprecipitating PHD2 / prolyl hydroxylase in SH-SY5Y treated with 0.1mM cobalt chloride for 8 hours whole cell lysate.
Lane 1 (input): SH-SY5Y (Human neuroblastoma epithelial cell) treated with 0.1mM cobalt chloride for 8 hours whole cell lysate 10µg
Lane 2 (+): ab133630 & SH-SY5Y treated with 0.1mM cobalt chloride for 8 hours whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab133630 in SH-SY5Y treated with 0.1mM cobalt chloride for 8 hours whole cell lysate
For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1:1000 dilution.
Blocking and diluting buffer: 5% NFDM/TBST.

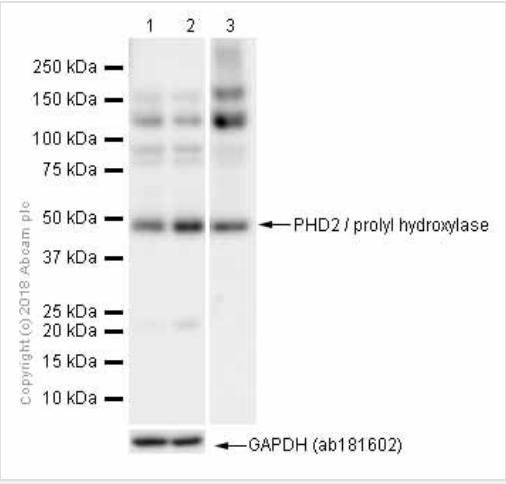


OI-RD Scanning - Anti-PHD2 / prolyl hydroxylase antibody [EPR3660(B)(2)] (ab133630)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D



Western blot - Anti-PHD2 / prolyl hydroxylase antibody [EPR3660(B)(2)] (ab133630)

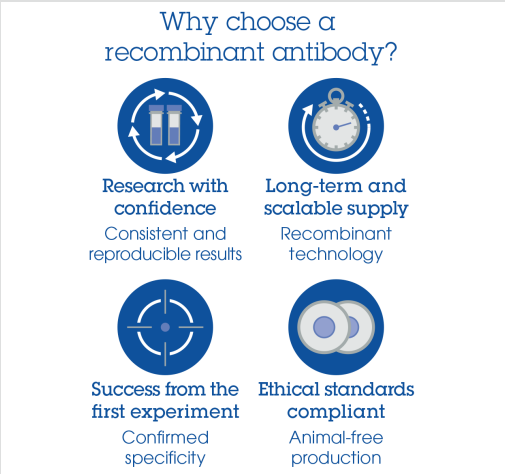
All lanes : Anti-PHD2 / prolyl hydroxylase antibody [EPR3660(B)(2)] (ab133630) at 1/5000 dilution (Purified)

Lane 1 : SH-SY5Y (Human neuroblastoma epithelial cell) whole cell lysates with 5% NFDM/TBST
Lane 2 : SH-SY5Y (Human neuroblastoma epithelial cell) treated with 0.1mM cobalt chloride for 8 hours whole cell lysates with 5% NFDM/TBST
Lane 3 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates with 5% NFDM/TBST

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 46 kDa
Observed band size: 46 kDa



Anti-PHD2 / prolyl hydroxylase antibody

[EPR3660(B)(2)] (ab133630)

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