

Product Specification Sheet

HIF-Prolyl hydroxylase 1-3 Antibodies

Cat. PHDC12-P	PHD1-3 (pan) control/blocking peptide # 1	SIZE: 100 ug
Cat. PHDC12-A	Rabbit Anti- PHD1-3 (pan)3 IgG # 1 (aff pure)	SIZE: 100 ug

Oxygen is absolutely critical for the survival of mammalian cells. Hypoxia induced factor (HIF) is a transcriptional complex that plays a central role in mammalian oxygen homeostasis. There are 3-types of alpha subunits (HIF1-3alpha) and one HIF-beta subunits. However, the three HIF-alpha subunits are regulated by oxygen in a similar fashion, i.e. by regulated stabilization of the alpha-subunits. Under normal conditions, HIF-alpha Prolines are hydroxylated at Pro-402 and Pro-564. This allows binding of von Hippel-Lindau (VHL), the substrate recognition component of the E3 ubiquitinated ligase complex, subsequent ubiquitination and degradation of HIF-alpha by the proteasome. Under hypoxic conditions, hydroxylation of HIF-alpha is inhibited and this prevents HIF-alpha degradation. The enzymes responsible for HIF-hydroxylation are known as HIF-prolyl hydroxylases (PHD1-3 or HPH1, HPH2, and HPH3). The three PHDs have been identified to hydroxylate the motif, LXXLAP* with *P being the hydroxyproline.

Source of Antigen and Antibodies

Antigen	18-aa peptide of Human PHD2/EGLN1 (1); Designated (PHDC12-P or control peptide). This peptide is significantly conserved in PHD1-3 in mouse, rat, and human.conjugated to KLH; Epitope location ~N-terminus
Ab Host/type	Rabbit, Polyclonal Aff pure IgG, (Cat # PHDC12-A) purified over the antigen column
2-Ab	Cat # 20320, goat anti-rabbit IgG-HRP (AP, biotin, FITC conjugates also available).
-ve control	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as -ve control

Form & Storage of Antibodies/Peptide Control

Affinity pure IgG
100 ug/100ul solution lyophilized powder
Supplied in **Buffer:** PBS+0.1% BSA
Reconstitute powder in PBS at 1mg/ml

Control/blocking peptide
100 ug/100 ul solution lyophilized powder
Supplied in Buffer: PBS pH 7.5,
Reconstitute powder in PBS at 1 mg/ml.

Storage

Short-term: unopened, undiluted liquid vials at -20°C and powder at 4°C or -20°C..

Long-term: at -20°C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at -20°C or below.

Shipping: 4°C for solutions and room temp for powder

Recommended Usage

Western Blotting 1-5 ug/ml for affinity pure using Chemiluminescence technique.

ELISA (1:10K-1:100K; using 50-100 ng of control peptide/well).

Histochemistry & Immunofluorescence: Not tested.

Specificity & Cross-reactivity

Human PHDC12-P was selected to represent a conserved region in PHD1-3 proteins. It is 100% conserved in mouse, rat, and human PHD2/EGLN1. It is 94% conserved in mouse, rat, and human PHD1/EGLN2. Homology with the mouse, rat, and human PHD3/EGLN3 is 88%. The antibodies are expected to recognize PHD1-3 protein from mouse, rat, and human. However, the antibody reactivity needs to be experimentally confirmed. Control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-10 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity

General References: trausberg RL et al (2002) PNAS 99, 16899-16903; Taylor MS et al (2001) Gene 275, 125-132; Epstein AC et al (2001) Cell 107, 43-54; Semenza GL et al (2001) Cell 107, 1-3; Bruick RK et al (2001) Science 294, 1337-1340; Oehme F et al (2002) BBRC 296, 343-349

*This product is for in vitro research use only.

Related material available from ADI

Antibodies to PHD1-3, HIF1-3, and other apoptosis proteins
Western Blot recycling kit (Use the same blot to probe with multiple antibodies NBC1-3)

PHDC12-A-P 70912J