Hydroxy-HIF-1α (Pro564) (D43B5) XP® Rabbit mAb



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Applications: W, IP, IF-IC	Reactivity: H Mk	Sensitivity: Endogenous	MW (kDa): 120	Source/Isotype: Rabbit IgG	UniProt ID: #Q16665	Entrez-Gene Id 3091	
Product Usage		Application Dilution					
Information		Western Blotting			1:1000		
		Immunoprecipitation			1:50		
		Immunofluorescence	(Immunocytochem	istry)	1:3200) - 1:6400	
Storage		Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 μ g/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. Do not aliquot the antibody.					
Specificity/Sensitivity		Hydroxy-HIF-1 α (Pro564) (D43B5) XP [®] Rabbit mAb detects endogenous levels of HIF-1 α only when hydroxylated at Pro564. This antibody may cross react with other overexpressed proline hydroxylated proteins.					
Species predicted to react based on 100% sequence homology		Mouse, Rat, Chicken, Xenopus, Zebrafish, Pig					
Source / Purification		Monoclonal antibody is produced by immunizing animals with a synthetic hydroxypeptide corresponding to residues surrounding Pro564 of human HIF-1 α .					
Background		Hypoxia-inducible factor 1 (HIF1) is a heterodimeric transcription factor that plays a critical role in the cellular response to hypoxia (1). The HIF1 complex consists of two subunits, HIF- 1α and HIF- 1β , which are basic helix-loop-helix proteins of the PAS (Per, ARNT, Sim) family (2). HIF1 regulates the transcription of a broad range of genes that facilitate responses to the hypoxic environment, including genes regulating angiogenesis, erythropoiesis, cell cycle, metabolism, and apoptosis. The widely expressed HIF- 1α is typically degraded rapidly in normoxic cells by the ubiquitin/proteasomal pathway. Under normoxic conditions, HIF- 1α is proline hydroxylated leading to a conformational change that promotes binding to the von Hippel-Lindau protein (VHL) E3 ligase complex; ubiquitination and proteasomal degradation follows (3,4). Both hypoxic conditions and chemical hydroxylase inhibitors (such as desferrioxamine and cobalt) inhibit HIF- 1α degradation and lead to its stabilization. In addition, HIF- 1α can be induced in an oxygen-independent manner by various cytokines through the PI3K-AKT-mTOR					

HIF-1β is also known as AhR nuclear translocator (ARNT) due to its ability to partner with the aryl hydrocarbon receptor (AhR) to form a heterodimeric transcription factor complex (8). Together with AhR, HIF-1β plays an important role in xenobiotics metabolism (8). In addition, a chromosomal translocation leading to a TEL-ARNT fusion protein is associated with acute myeloblastic leukemia (9). Studies also found that ARNT/HIF-1 β expression levels decrease significantly in pancreatic islets from patients with type 2 diabetes, suggesting that HIF-1β plays an important role in pancreatic β-cell function (10).~Two critical prolines in HIF-1α (Pro564 and Pro402) can be hydroxylated by proline hydroxylase under normoxia conditions. Hydroxylation of HIF-1α leads to its binding to VHL and ubiquitin mediated degradation (3,11,12).

Background References

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pathway (5-7).

- 4. Maxwell, P.H. et al. (1999) Nature 399, 271-5.
- 5. Fukuda, R. et al. (2002) *J Biol Chem* 277, 38205-11.
- 6. Jiang, B.H. et al. (2001) Cell Growth Differ 12, 363-9.
- 7. Laughner, E. et al. (2001) *Mol Cell Biol* 21, 3995-4004.
- 8. Walisser, J.A. et al. (2004) Proc Natl Acad Sci U S A 101, 16677-82.
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- 10. Gunton, J.E. et al. (2005) Cell 122, 337-49.
- 11. Ivan, M. et al. (2001) Science 292, 464-8.
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Species Reactivity Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer IMPORTANT: For western blots, incubate membrane with diluted primary antibody in 5% w/v BSA, 1X

TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

Applications Key W: Western Blotting **IP**: Immunoprecipitation **IF-IC**: Immunofluorescence (Immunocytochemistry)

Cross-Reactivity Key H: Human Mk: Monkey

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