Recombinant PHD2 (EGLN1) protein

Catalog No: 81065, 81765
Lot No: 24717001
Expressed In: Baculovirus
Quantity: 20, 1000 µg
Concentration: 0.2 µg/µl
Source: Human

Buffer Contents: Recombinant PHD2 / EGLN1 protein is supplied in 25 mM HEPES-NaOH pH 7.5, 300 mM NaCl, 10% glycerol, 0.04% Triton X-100 and 0.5 mM TCEP.

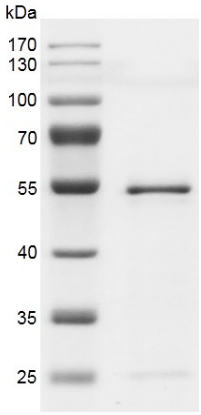
Background: Prolyl Hydroxylase Domain-Containing Protein 2 (PHD2) or Egl-9 Family Hypoxia Inducible Factor 1 (EGLN1), also known as HIF-PH2 or HPH-2, is a prolyl hydroxylase. PHD3 is a cellular oxygen sensor that catalyzes the post-translational formation of 4-hydroxyproline in hypoxia-inducible factor (HIF) alpha proteins under normoxic conditions. It can hydroxylate a specific proline found in each of the oxygen-dependent degradation (ODD) domains (N-terminal, NODD, and C-terminal, CODD) of HIF1A, also HIF2A. PHD1 has a preference for the CODD site for both HIF1A and HIF2A. Hydroxylated HIFs are targeted for proteasomal degradation via the von Hippel-Lindau ubiquitination complex. Under hypoxic conditions, the hydroxylation reaction is attenuated allowing HIFs to escape degradation resulting in their translocation to the nucleus, heterodimerization with HIF1B, and increased expression of hypoxy-inducible genes. PHD2 is the most important isozyme under normoxia and, through regulating the stability of HIF1, involved in various hypoxia-influenced processes such as angiogenesis in retinal and cardiac functionality. Its target proteins are preferentially recognized via a LXXLAP motif.

Protein Details: Recombinant human PHD2 (EGLN1) protein was expressed in a baculovirus expression system as the full length protein (accession number NP_071334.1) with an N-terminal FLAG tag. The molecular weight of the protein is 47 kDa.

Application Notes: This protein is suitable for use in binding assays, inhibitor screening, and selectivity profiling.

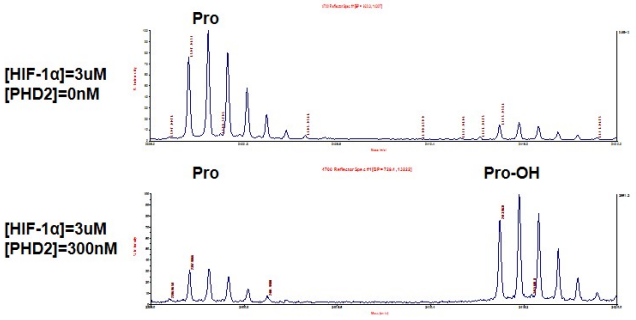
Assay Conditions: 3 µM HIF1A (HIF-1α) peptide (DLDLEALAPYIPADDDFQL) was incubated with 300 nM PHD2 protein in 30 µl reaction system containing 20 mM Tris-HCl pH 7.5, 5 mM KCl, 1.5 mM MgCl2, 1 mM DTT, 100 µM 2-oxoglutarate, 100 µM ascorbate and 50 µM (NH4)2Fe(SO4)2·6H2O for 2 hours at 30°C. MALDI-TOF was used for detection.

Storage and Guarantee: Recombinant proteins in solution are temperature sensitive and must be stored at -80°C to prevent degradation. Avoid repeated freeze/thaw cycles and keep on ice when not in storage. This product is for research use only and is not for use in diagnostic procedures. This product is guaranteed for 6 months from date of arrival.



Recombinant PHD2 / EGLN1 protein gel

10% SDS-PAGE Coomassie staining
MW: 47 kDa
Purity: >90%



MALDI-TOF for PHD2 / EGLN1 protein

3 μM HIF1A peptide was incubated with 300 nM PHD2 protein in 30 μl reaction system for 2 hours at 30°C. The reaction product was detected by MALDI-TOF. Single 3 μM HIF1-α peptide was used as negative control.