SANTA CRUZ BIOTECHNOLOGY, INC.

HIF PHD2 siRNA (h): sc-45537

BACKGROUND

Prolyl hydroxylase domain proteins HIF PHD1, HIF PHD2 and HIF PHD3 (known as PHD1, PHD2 and PHD3 in rodents, respectively) can hydroxylate HIF-α subunits. Hypoxia-inducible factor (HIF) is a transcriptional regulator important in several aspects of oxygen homeostasis. The prolyl hydroxylases catalyze the posttranslational formation of 4-hydroxyproline in HIF-α proteins. HIF PHD1, which is widely expressed, with highest levels of expression in testis, functions as a cellular oxygen sensor and is important in cell growth regulation. HIF PHD1 can localize to the nucleus or the cytoplasm and is also detected in hormone responsive tissues, such as normal and cancerous mammary, ovarian and prostate epithelium. HIF PHD1 is encoded by EGLN2, which maps to chromosome 19q13.2. HIF PHD2 is regarded as the main cellular oxygen sensor, as RNA interference against HIF PHD2, but not HIF PHD1 or HIF PHD3, is enough to stabilize HIF-1α in normoxia. HIF PHD2, a direct HIF target gene, is expressed mainly in skeletal muscle, heart, kidney and brain. HIF PHD3 may play a role in the regulation of cell growth in muscle cells and in apoptosis in neuronal tissue. HIF PHD3 is widely expressed, although the highest levels can be detected in placenta and heart.

CHROMOSOMAL LOCATION

Genetic locus: EGLN1 (human) mapping to 1q42.2.

PRODUCT

HIF PHD2 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HIF PHD2 shRNA Plasmid (h): sc-45537-SH and HIF PHD2 shRNA (h) Lentiviral Particles: sc-45537-V as alternate gene silencing products.

For independent verification of HIF PHD2 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-45537A, sc-45537B and sc-45537C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

HIF PHD2 siRNA (h) is recommended for the inhibition of HIF PHD2 expression in human cells.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

HIF PHD2 (H-8): sc-271835 is recommended as a control antibody for monitoring of HIF PHD2 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HIF PHD2 gene expression knockdown using RT-PCR Primer: HIF PHD2 (h)-PR: sc-45537-PR (20 µl, 506 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Soeda, A., et al. 2009. Hypoxia promotes expansion of the CD133-positive glioma stem cells through activation of HIF-1α. Oncogene 28: 3949-3959.
2. Bienes-Martínez, R., et al. 2012. Autocrine stimulation of clear-cell renal carcinoma cell migration in hypoxia via HIF-independent suppression of thrombospondin-1. Sci. Rep. 2: 788.
3. Hou, P., et al. 2014. Intermediary metabolite precursor dimethyl-2-ketoglutarate stabilizes hypoxia-inducible factor-1α by inhibiting prolyl-4-hydroxylase PHD2. PLoS ONE 9: e113865.
4. Labrousse-Arias, D., et al. 2017. VHL promotes immune response against renal cell carcinoma via NFκB-dependent regulation of VCAM-1. J. Cell Biol. 216: 835-847.
5. Bora-Singhal, N., et al. 2022. A novel PHD2/VHL-mediated regulation of YAP1 contributes to VEGF expression and angiogenesis. Cancer Res. Commun. 2: 624-638.
6. Zheng, J., et al. 2023. Overactivated NRF2 induces pseudohypoxia in hepatocellular carcinoma by stabilizing HIF-1α. Free Radic. Biol. Med. 194: 347-356.
7. Guillen-Quispe, Y.N., et al. 2024. Non-canonical function of prolyl hydroxylase domain 2 in breast cancer cell growth and progression: role of peptidyl-prolyl *cis-trans* isomerase NIMA-interacting 1. J. Cancer Prev. 29: 129-139.

RESEARCH USE

For research use only, not for use in diagnostic procedures.