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HIF-1 α siRNA (m2): sc-44308

BACKGROUND

Cell growth and viability is compromised by oxygen deprivation (hypoxia). Hypoxia-inducible factors, including HIF-1 α , HIF-1 β (also designated Arnt 1), EPAS-1 (also designated HIF-2 α) and HIF-3 α , induce glycolysis, erythropoiesis and angiogenesis in order to restore oxygen homeostasis. Hypoxia-inducible factors are members of the Per-Arnt-Sim (PAS) domain transcription factor family. In response to hypoxia, HIF-1 α is upregulated and forms a heterodimer with Arnt 1 to form the HIF-1 complex. The HIF-1 complex recognizes and binds to the hypoxia responsive element (HRE) of hypoxia-inducible genes, thereby activating transcription. Hypoxia-inducible expression of some genes such as Glut-1, p53, p21 or Bcl-2, is HIF-1 α dependent, whereas expression of others, such as p27, GADD 153 or HO-1, is HIF-1 α independent. EPAS-1 and HIF-3 α have also been shown to form heterodimeric complexes with Arnt 1 in response to hypoxia.

REFERENCES

1. Wang, G.L., et al. 1995. Hypoxia-inducible factor 1 is a basic-helix-loop-helix-PAS heterodimer regulated by cellular O₂ tension. *Proc. Natl. Acad. Sci. USA* 92: 5510-5514.
2. Tian, H., et al. 1997. Endothelial PAS domain protein 1 (EPAS1), a transcription factor selectively expressed in endothelial cells. *Genes Dev.* 11: 72-82.
3. Luo, G., et al. 1997. Molecular characterization of the murine HIF-1 α locus. *Gene Expr.* 6: 287-299.

CHROMOSOMAL LOCATION

Genetic locus: Hif1a (mouse) mapping to 12 C3.

PRODUCT

HIF-1 α siRNA (m2) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HIF-1 α shRNA Plasmid (m2): sc-44308-SH and HIF-1 α shRNA (m2) Lentiviral Particles: sc-44308-V as alternate gene silencing products.

For independent verification of HIF-1 α (m2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44308A, sc-44308B and sc-44308C.

STORAGE AND RESUSPENSION

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

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

APPLICATIONS

HIF-1 α siRNA (m2) is recommended for the inhibition of HIF-1 α expression in mouse cells.

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 μ M in 66 μ l. 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-1 α (28b): sc-13515 is recommended as a control antibody for monitoring of HIF-1 α 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).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HIF-1 α gene expression knockdown using RT-PCR Primer: HIF-1 α (m2)-PR: sc-44308-PR (20 μ l, 521 bp). Annealing temperature for the primers should be 55-60 $^{\circ}$ C and the extension temperature should be 68-72 $^{\circ}$ C.

SELECT PRODUCT CITATIONS

1. Singh, A.K., et al. 2012. Intracellular pathogen *Leishmania donovani* activates hypoxia inducible factor-1 by dual mechanism for survival advantage within macrophage. *PLoS ONE* 7: e38489.
2. Khan, M., et al. 2015. Promoting endothelial function by S-nitrosoglutathione through the HIF-1 α /VEGF pathway stimulates neurorepair and functional recovery following experimental stroke in rats. *Drug Des. Devel. Ther.* 9: 2233-2247.
3. Ambade, A., et al. 2016. Hepatocellular carcinoma is accelerated by NASH involving M2 macrophage polarization mediated by HIF-1 α induced IL-10. *Oncoimmunology* 5: e1221557.

RESEARCH USE

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