

# HIF-1 $\alpha$ siRNA (m): sc-35562

## BACKGROUND

Cell growth and viability is compromised by oxygen deprivation (hypoxia). Hypoxia-inducible factors, including HIF-1 $\alpha$ , Arnt 1 (also designated HIF-1 $\beta$ ), EPAS-1 (also designated HIF-2 $\alpha$ ) and HIF-3 $\alpha$ , 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 $\alpha$  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 $\alpha$  dependent, whereas expression of others, such as p27, GADD 153 or HO-1, is HIF-1 $\alpha$  independent. EPAS-1 and HIF-3 $\alpha$  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 O<sub>2</sub> tension. *Proc. Natl. Acad. Sci. USA* 92: 5510-5514.
2. Tian, H., et al. 1997. Endothelial PAS domain protein 1 (EPAS-1), 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 $\alpha$  locus. *Gene Expr.* 6: 287-299.
4. Carmeliet, P., et al. 1998. Role of HIF-1 $\alpha$  in hypoxia-mediated apoptosis, cell proliferation and tumour angiogenesis. *Nature* 394: 485-490.

## CHROMOSOMAL LOCATION

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

## PRODUCT

HIF-1 $\alpha$  siRNA (m) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HIF-1 $\alpha$  shRNA Plasmid (m): sc-35562-SH and HIF-1 $\alpha$  shRNA (m) Lentiviral Particles: sc-35562-V as alternate gene silencing products.

For independent verification of HIF-1 $\alpha$  (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-35562A, sc-35562B and sc-35562C.

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

## APPLICATIONS

HIF-1 $\alpha$  siRNA (m) is recommended for the inhibition of HIF-1 $\alpha$  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  $\mu$ M in 66  $\mu$ 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 $\alpha$  (28b): sc-13515 is recommended as a control antibody for monitoring of HIF-1 $\alpha$  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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HIF-1 $\alpha$  gene expression knockdown using RT-PCR Primer: HIF-1 $\alpha$  (h)-PR: sc-35561-PR (20  $\mu$ l, 457 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Lee, S.J., et al. 2011. Beclin 1 deficiency is associated with increased hypoxia-induced angiogenesis. *Autophagy* 7: 829-839.
2. Yang, Z., et al. 2014. Hypoxia induces autophagic cell death through hypoxia-inducible factor 1 $\alpha$  in microglia. *PLoS ONE* 9: e96509.

## RESEARCH USE

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

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.