

KLF8 siRNA (h): sc-75390

BACKGROUND

The Krüppel-type zinc finger transcription factors comprise a conserved family of DNA binding proteins that are important in developmental regulation. The Krüppel zinc finger transcription factor was initially identified in *Drosophila* as a segmentation gene. Krüppel-like factor 8 (KLF8), also called basic Krüppel-like factor 3 and zinc finger protein 741, is a 359 amino acid transcriptional repressor that binds CACCC elements in DNA and activates or represses their target genes in a context-dependent manner. KLF8 is expressed ubiquitously in the nucleus of many cell types and its expression is elevated in several human cancers. KLF8 is post-translationally modified and negatively regulated by sumoylation via SUMO-1, SUMO-2 or SUMO-3. Mutation of the sumoylation site, Lysine 67, to Arginine 67 enhances the ability of KLF8 to repress or activate its target promoters.

REFERENCES

1. van Vliet, J., et al. 2000. Human Krüppel-like factor 8: a CACCC-box binding protein that associates with CtBP and represses transcription. *Nucleic Acids Res.* 28: 1955-1962.
2. Lossi, A.M., et al. 2002. Abnormal expression of the KLF8 (ZNF741) gene in a female patient with an X; autosome translocation t(X;21)(p11.2;q22.3) and non-syndromic mental retardation. *J. Med. Genet.* 39: 113-117.
3. Zhao, J., et al. 2003. Identification of transcription factor KLF8 as a downstream target of focal adhesion kinase in its regulation of cyclin D1 and cell cycle progression. *Mol. Cell* 11: 1503-1515.
4. Chiambaretta, F., et al. 2004. Cell and tissue specific expression of human Krüppel-like transcription factors in human ocular surface. *Mol. Vis.* 10: 901-909.
5. Cox, B.D., et al. 2006. New concepts regarding focal adhesion kinase promotion of cell migration and proliferation. *J. Cell. Biochem.* 99: 35-52.
6. Wei, H., et al. 2006. Sumoylation delimits KLF8 transcriptional activity associated with the cell cycle regulation. *J. Biol. Chem.* 281: 16664-16671.

CHROMOSOMAL LOCATION

Genetic locus: KLF8 (human) mapping to Xp11.21.

PRODUCT

KLF8 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see KLF8 shRNA Plasmid (h): sc-75390-SH and KLF8 shRNA (h) Lentiviral Particles: sc-75390-V as alternate gene silencing products.

For independent verification of KLF8 (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-75390A, sc-75390B and sc-75390C.

PROTOCOLS

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

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 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

KLF8 siRNA (h) is recommended for the inhibition of KLF8 expression in human 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

KLF8 (12D2): sc-134375 is recommended as a control antibody for monitoring of KLF8 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 KLF8 gene expression knockdown using RT-PCR Primer: KLF8 (h)-PR: sc-75390-PR (20 μ l, 600 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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