

GLI-2 siRNA (m): sc-145421

BACKGROUND

Zinc-finger proteins contain DNA-binding domains and have a wide variety of functions, most of which encompass some form of transcriptional activation or repression. The majority of zinc-finger proteins contain a Krüppel-type DNA binding domain and a KRAB domain, which is thought to interact with KAP1, thereby recruiting histone modifying proteins. GLI-2 (GLI family zinc finger 2), also known as HPE9 or THP (tax helper protein), is a 1,586 amino acid nuclear protein that acts as a transcriptional activator and belongs to the GLI C₂H₂-type zinc-finger protein family. Localized to the nucleus, GLI-2 is thought to play a role in embryogenesis. The gene encoding GLI-2 maps to human chromosome 2q14.2, and when defective is the cause of holoprosencephaly type 9 (HPE9). GLI-2 exists as five alternatively spliced isoforms.

REFERENCES

1. Ruppert, J.M., et al. 1988. The GLI-Krüppel family of human genes. *Mol. Cell. Biol.* 8: 3104-3113.
2. Tanimura, A., et al. 1993. A new regulatory element that augments the Tax-dependent enhancer of human T-cell leukemia virus type 1 and cloning of cDNAs encoding its binding proteins. *J. Virol.* 67: 5375-5382.
3. Tanimura, A., et al. 1998. Cloning of novel isoforms of the human GLI-2 oncogene and their activities to enhance tax-dependent transcription of the human T-cell leukemia virus type 1 genome. *J. Virol.* 72: 3958-3964.
4. Roessler, E., et al. 2003. Loss-of-function mutations in the human GLI-2 gene are associated with pituitary anomalies and holoprosencephaly-like features. *Proc. Natl. Acad. Sci. USA* 100: 13424-13429.
5. Roessler, E., et al. 2005. A previously unidentified amino-terminal domain regulates transcriptional activity of wild-type and disease-associated human GLI-2. *Hum. Mol. Genet.* 14: 2181-2188.
6. Filion, G.J., et al. 2006. A family of human zinc finger proteins that bind methylated DNA and repress transcription. *Mol. Cell. Biol.* 26: 169-181.

CHROMOSOMAL LOCATION

Genetic locus: Gli2 (mouse) mapping to 1 E2.3.

PRODUCT

GLI-2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GLI-2 shRNA Plasmid (m): sc-145421-SH and GLI-2 shRNA (m) Lentiviral Particles: sc-145421-V as alternate gene silencing products.

For independent verification of GLI-2 (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-145421A, sc-145421B and sc-145421C.

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

GLI-2 siRNA (m) is recommended for the inhibition of GLI-2 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

GLI-2 (C-10): sc-271786 is recommended as a control antibody for monitoring of GLI-2 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 GLI-2 gene expression knockdown using RT-PCR Primer: GLI-2 (m)-PR: sc-145421-PR (20 μ l, 565 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.