SANTA CRUZ BIOTECHNOLOGY, INC.

glypican-3 siRNA (m): sc-40641



BACKGROUND

Glypican-3 (GPC3) is an integral membrane protein that is mutated in the Simpson-Golabi-Behmel syndrome (SGBS). SGBS is characterized by preand post-natal overgrowth and is a recessive X-linked condition. Glypican-3, also designated OCI-5 in rat, is a member of the glypican family of heparan sulfate proteoglycans, which attach to the cell membrane via a glycosyl-phosphatidyl-inositol (GPI) anchor. Expression of glypican-3 is detected in embryonic mesodermal lung, liver and kidney tissues. Glypican-3 is thought to regulate tissue and organ growth through interactions with growth factors such as Insulin-like growth factor-II (IGF-II) or fibroblast growth factor-2 (FGF-2). Glypican-3 may be downregulated by various means, including promoter hypermethylation or the repression of specific transcription factors.

REFERENCES

- 1. Pilia, G., et al. 1996. Mutations in GPC3, a glypican gene, cause the Simpson-Golabi-Behmel overgrowth syndrome. Nat. Genet. 12: 241-247.
- Song, H.H., et al. 1997. OCI-5/rat glypican-3 binds to fibroblast growth factor-2 but not to Insulin-like growth factor-2. J. Biol. Chem. 272: 7574-7577.
- Li, M., et al. 1997. Expression of OCI-5/glypican-3 during intestinal morphogenesis: regulation by cell shape in intestinal epithelial cells. Exp. Cell Res. 235: 3-12.
- Gonzalez, A.D., et al. 1998. OCI-5/GPC3, a glypican encoded by a gene that is mutated in the Simpson-Golabi-Behmel overgrowth syndrome, induces apoptosis in a cell line-specific manner. J. Cell Biol. 141: 1407-1414.
- 5. Cano-Gauci, D.F., et al. 1999. Glypican-3-deficient mice exhibit developmental overgrowth and some of the abnormalities typical of Simpson-Golabi-Behmel syndrome. J. Cell Biol. 146: 255-264.
- 6. Lin, H., et al. 1999. Frequent silencing of the GPC3 gene in ovarian cancer cell lines. Cancer Res. 59: 807-810.
- Murthy, S.S., et al. 2000. Expression of GPC3, an X-linked recessive overgrowth gene, is silenced in malignant mesothelioma. Oncogene 19: 410-416.

CHROMOSOMAL LOCATION

Genetic locus: Gpc3 (mouse) mapping to X A5.

PRODUCT

glypican-3 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 glypican-3 shRNA Plasmid (m): sc-40641-SH and glypican-3 shRNA (m) Lentiviral Particles: sc-40641-V as alternate gene silencing products.

For independent verification of glypican-3 (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-40641A, sc-40641B and sc-40641C.

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

glypican-3 siRNA (m) is recommended for the inhibition of glypican-3 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

glypican-3 (F-3): sc-390587 is recommended as a control antibody for monitoring of glypican-3 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 MarkerTM 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 glypican-3 gene expression knockdown using RT-PCR Primer: glypican-3 (m)-PR: sc-40641-PR (20 μ I). 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.

PROTOCOLS

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