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glypican-2 siRNA (h): sc-44386

BACKGROUND

Glypican-1 (GPC1), glypican-2 (GPC2) and glypican-3 (GPC3) are members of the glypican family of heparan sulfate proteoglycans, which attach to the cell membrane via a glycosylphosphatidylinositol (GPI) anchor. Cell-surface heparan sulfate proteoglycans participate in molecular events that regulate cell adhesion, migration, and proliferation. Glypican-2, a cell surface proteoglycan bearing heparan sulfate, may have a function related to the motile behaviors of developing neurons. Ligation of cell-surface glypican-2 with midkine (MK) or an antibody against epitope-tagged glypican-2 induces cell adhesion and promotes neurite outgrowth. MK binds to heparan sulfate chains of glypican-2 in a manner similar to syndecan-3, but different localization of epitope-tagged glypican-2 and syndecan-3 on the surface of N2 α cells suggests that they may play different roles in MK-mediated neural function.

REFERENCES

1. Stipp, C.S. et al. 1994. Cerebroglycan: an integral membrane heparan sulfate proteoglycan that is unique to the developing nervous system and expressed specifically during neuronal differentiation. *J. Cell Biol.* 124: 149-160.
2. 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.
3. 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.
4. 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.
5. Kurosawa, N. et al. 2001. Glypican-2 binds to midkine: the role of glypican-2 in neuronal cell adhesion and neurite outgrowth. *Glycoconj. J.* 18: 499-507.

CHROMOSOMAL LOCATION

Genetic locus: GPC2 (human) mapping to 7q22.1.

PRODUCT

glypican-2 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 glypican-2 shRNA Plasmid (h): sc-44386-SH and glypican-2 shRNA (h) Lentiviral Particles: sc-44386-V as alternate gene silencing products.

For independent verification of glypican-2 (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-44386A, sc-44386B and sc-44386C.

PROTOCOLS

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

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

glypican-2 siRNA (h) is recommended for the inhibition of glypican-2 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

glypican-2 (F-5): sc-393824 is recommended as a control antibody for monitoring of glypican-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 λ BP-HRP: sc-516132 or m-IgG λ BP-HRP (Cruz Marker): sc-516132-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 λ BP-FITC: sc-516185 or m-IgG λ BP-PE: sc-516186 (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-2 gene expression knockdown using RT-PCR Primer: glypican-2 (h)-PR: sc-44386-PR (20 μ l). Annealing temperature for the primers should be 55-60 $^{\circ}$ C and the extension temperature should be 68-72 $^{\circ}$ C.

RESEARCH USE

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