

GFR α -1 siRNA (h): sc-35469

BACKGROUND

Glial cell line-derived neurotrophic factor (GDNF) and the related neurotrophic factor neurturin (NTN) are potent survival factors for central and peripheral neurons. GDNF is a glycosylated, disulfide-bonded homodimer that is distantly related to the TGF β superfamily of growth factors. Three receptors for these factors, GFR α -1 (also designated GDNFR- α , RETL1 or TrnR-1), GFR α -2 (also designated GDNFR- β , RETL2, NTNR- α or TrnR-2) and GFR α -3 have been identified. The receptors do not contain transmembrane domains and are attached to the cell membrane by glycosyl-phosphoinositol linkage. Both GFR α -1 and GFR α -2 have been shown to mediate the GDNF-dependent and NTN-dependent phosphorylation and activation of the tyrosine kinase Ret. GFR α -3 is expressed only during development.

REFERENCES

1. Lin, L.F., et al. 1993. GDNF: a glial cell line-derived neurotrophic factor for midbrain dopaminergic neurons. *Science* 260: 1130-1132.
2. Jing, S., et al. 1996. GDNF-induced activation of the Ret protein tyrosine kinase is mediated by GDNFR- α , a novel receptor for GDNF. *Cell* 85: 1113-1124.
3. Treanor, J.J., et al. 1996. Characterization of a multi-component receptor for GDNF. *Nature* 382: 80-83.
4. Kotzbauer, P.T., et al. 1996. Neurturin, a relative of glial-cell-line-derived neurotrophic factor. *Nature* 384: 467-470.
5. Baloh, R.H., et al. 1997. TrnR2, a novel receptor that mediates neurturin and GDNF signaling through Ret. *Neuron* 18: 793-802.

CHROMOSOMAL LOCATION

Genetic locus: GFRA1 (human) mapping to 10q25.3.

PRODUCT

GFR α -1 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 GFR α -1 shRNA Plasmid (h): sc-35469-SH and GFR α -1 shRNA (h) Lentiviral Particles: sc-35469-V as alternate gene silencing products.

For independent verification of GFR α -1 (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-35469A, sc-35469B and sc-35469C.

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

GFR α -1 siRNA (h) is recommended for the inhibition of GFR α -1 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

GFR α -1 (E-11): sc-271546 is recommended as a control antibody for monitoring of GFR α -1 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 GFR α -1 gene expression knockdown using RT-PCR Primer: GFR α -1 (h)-PR: sc-35469-PR (20 μ l, 526 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Kim, M., et al. 2017. GFRA1 promotes cisplatin-induced chemoresistance in osteosarcoma by inducing autophagy. *Autophagy* 13: 149-168.
2. Ban, K., et al. 2017. Ret signaling in prostate cancer. *Clin. Cancer Res.* 23: 4885-4896.
3. Liu, D., et al. 2022. σ -1 receptor activation alleviates blood-brain barrier disruption post cerebral ischemia stroke by stimulating the GDNF-GFR α 1-RET pathway. *Exp. Neurol.* 347: 113867.

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.