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IGF-IR α/β siRNA (r): sc-270198

BACKGROUND

Receptor tyrosine kinases (RTKs) are transmembrane molecular scaffolds that influence cellular processes including the cell cycle, cell migration, cell metabolism, cell survival, proliferation and differentiation. Insulin-like growth factor-I receptor (IGF-IR) is an RTK that stimulates growth in many different cell types, blocks apoptosis, acts as an intermediate of many growth hormone responses and may stimulate the growth of some types of cancer. The IGF-IR cognate ligand Insulin-like growth factor-I (IGF-I) promotes association of IGF-IR with Shc, GRB2 and Sos 1, which initiates Ras and ERK kinase cascades, thereby modifying transcription factor activity, such as activation of the Elk transcription factors. The modular phosphotyrosine binding (PTB) domains of Insulin receptor substrate (IRS)-1 and -2 can associate with active IGF-IR and initiate phosphatidylinositol 3-kinase-dependent downstream signals. The human IGF-IR gene maps to chromosome 15q26.3 and encodes a 1,376 amino acid precursor protein that cleaves into α and β subunits. The human IGF-IR gene maps to chromosome 6q26 and encodes a 2,491 amino acid transmembrane protein.

REFERENCES

1. Frattali, A.L., et al. 1993. Molecular defects of Insulin/IGF-I receptor transmembrane signaling. *Ann. N.Y. Acad. Sci.* 687: 77-89.
2. Keller, S.R., et al. 1993. Insulin and IGF-I signaling through the Insulin receptor substrate-1. *Mol. Reprod. Dev.* 35: 346-352.
3. De Meyts, P., et al. 1995. Mechanism of Insulin and IGF-I receptor activation and signal transduction specificity. Receptor dimer cross-linking, bell-shaped curves, and sustained versus transient signaling. *Ann. N.Y. Acad. Sci.* 766: 388-401.
4. Song, R.X., et al. 2004. The role of Shc and Insulin-like growth factor 1 receptor in mediating the translocation of estrogen receptor α to the plasma membrane. *Proc. Natl. Acad. Sci. USA* 101: 2076-2081.
5. Mitsiades, C.S., et al. 2004. Inhibition of the Insulin-like growth factor receptor-1 tyrosine kinase activity as a therapeutic strategy for multiple myeloma, other hematologic malignancies, and solid tumors. *Cancer Cell* 5: 221-230.

CHROMOSOMAL LOCATION

Genetic locus: Igf1r (rat) mapping to 1q22.

PRODUCT

IGF-IR α/β siRNA (r) 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 IGF-IR α/β shRNA Plasmid (r): sc-270198-SH and IGF-IR α/β shRNA (r) Lentiviral Particles: sc-270198-V as alternate gene silencing products.

For independent verification of IGF-IR α/β (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270198A, sc-270198B and sc-270198C.

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

IGF-IR α/β siRNA (r) is recommended for the inhibition of IGF-IR α/β expression in rat 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

IGF-IR α (G-5): sc-271606 is recommended as a control antibody for monitoring of IGF-IR α/β 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 IGF-IR α/β gene expression knockdown using RT-PCR Primer: IGF-IR α/β (r)-PR: sc-270198-PR (20 μ l). 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.