

γ -FR siRNA (h): sc-39975

BACKGROUND

Folate is an essential vitamin that must be obtained from food intake through intestinal absorption in mammals. Folate and reduced folic acid derivatives bind to the folate receptor (FR) family, which mediates the endocytosis of 5-methyltetrahydrofolate into the cell. The folate receptors consist of five members, α , β , γ , γ' (which is produced by alternative splicing) and δ . α -FR and β -FR are attached to the membrane by a GPI anchor and are expressed in malignant tissues of epithelial and nonepithelial origin, respectively. γ -FR is expressed in tissues of hematopoietic origin, such as spleen, thymus and bone marrow, but the expression pattern of δ -FR is elusive, which suggests that it is highly restricted both spatially and temporally. α -FR is used as a highly selective tumor marker and may be targeted for the delivery of therapeutic compounds to tumor cells by coupling to derivatives of folic acid.

REFERENCES

1. Prasad, P.D., et al. 1994. Selective expression of the high-affinity isoform of the folate receptor (FR- α) in the human placental syncytiotrophoblast and choriocarcinoma cells. *Biochim. Biophys. Acta* 1223: 71-75.
2. Shen, F., et al. 1995. Folate receptor type γ is primarily a secretory protein due to lack of an efficient signal for glycosylphosphatidylinositol modification: protein characterization and cell type specificity. *Biochemistry* 34: 5660-5665.
3. Wang, H., et al. 1998. Structure and regulation of a polymorphic gene encoding folate receptor type γ/γ' . *Nucleic Acids Res.* 26: 2132-2142.
4. Said, H.M., et al. 2000. Adaptive regulation of intestinal folate uptake: effect of dietary folate deficiency. *Am. J. Physiol., Cell Physiol.* 279: C1889-C1895.
5. Spiegelstein, O., et al. 2000. Identification of two putative novel folate receptor genes in humans and mouse. *Gene* 258: 117-125.
6. Wang, H., et al. 2000. Differentiation-independent retinoid induction of folate receptor type β , a potential tumor target in myeloid leukemia. *Blood* 96: 3529-3536.
7. Sudimack, J., et al. 2000. Targeted drug delivery via the folate receptor. *Adv. Drug Deliv. Rev.* 41: 147-162.

CHROMOSOMAL LOCATION

Genetic locus: FOLR3 (human) mapping to 11q13.4.

PRODUCT

γ -FR 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 γ -FR shRNA Plasmid (h): sc-39975-SH and γ -FR shRNA (h) Lentiviral Particles: sc-39975-V as alternate gene silencing products.

For independent verification of γ -FR (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-39975A, sc-39975B and sc-39975C.

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

γ -FR siRNA (h) is recommended for the inhibition of γ -FR 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

FR (E-11): sc-515521 is recommended as a control antibody for monitoring of γ -FR 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 γ -FR gene expression knockdown using RT-PCR Primer: γ -FR (h)-PR: sc-39975-PR (20 μ l, 491 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.

PROTOCOLS

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