

RNFT1 siRNA (h): sc-93732

BACKGROUND

The RING-type zinc finger motif is present in a number of viral and eukaryotic proteins and is made of a conserved cysteine-rich domain that is able to bind two zinc atoms. Proteins that contain this conserved domain are generally involved in the ubiquitination pathway of protein degradation. RNFT1 (RING finger and transmembrane domain-containing protein 1), also known as PTD016, is a 435 amino acid multi-pass membrane protein that contains one RING-type zinc finger and exists as three alternatively spliced isoforms. The gene encoding RNFT1 maps to human chromosome 17, which comprises over 2.5% of the human genome and encodes over 1,200 genes. Two key tumor suppressor genes are associated with chromosome 17, namely, p53 and BRCA1. Malfunction or loss of p53 expression is associated with malignant cell growth and Li-Fraumeni syndrome. Like p53, BRCA1 is directly involved in DNA repair, and is linked to predisposition of cancers of the ovary, colon, prostate gland and fallopian tubes.

REFERENCES

1. Hall, J.M., et al. 1992. Closing in on a breast cancer gene on chromosome 17q. *Am. J. Hum. Genet.* 50: 1235-1242.
2. Freemont, P.S. 1993. The RING finger. A novel protein sequence motif related to the zinc finger. *Ann. N.Y. Acad. Sci.* 684: 174-192.
3. Borden, K.L. and Freemont, P.S. 1996. The RING finger domain: a recent example of a sequence-structure family. *Curr. Opin. Struct. Biol.* 6: 395-401.
4. Varley, J.M., et al. 1997. A detailed study of loss of heterozygosity on chromosome 17 in tumours from Li-Fraumeni patients carrying a mutation to the TP53 gene. *Oncogene* 14: 865-871.

CHROMOSOMAL LOCATION

Genetic locus: RNFT1 (human) mapping to 17q23.1.

PRODUCT

RNFT1 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 RNFT1 shRNA Plasmid (h): sc-93732-SH and RNFT1 shRNA (h) Lentiviral Particles: sc-93732-V as alternate gene silencing products.

For independent verification of RNFT1 (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-93732A, sc-93732B and sc-93732C.

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

RNFT1 siRNA (h) is recommended for the inhibition of RNFT1 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

RNFT1 (10G3): sc-134430 is recommended as a control antibody for monitoring of RNFT1 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 RNFT1 gene expression knockdown using RT-PCR Primer: RNFT1 (h)-PR: sc-93732-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.