SANTA CRUZ BIOTECHNOLOGY, INC.

TRIM35 siRNA (h): sc-76748

BACKGROUND

TRIM35 (tripartite motif-containing 35), also known as HLS5 (hemopoietic lineage switch protein 5) or MAIR (macrophage-derived apoptosis-inducing RBCC protein), is a widely expressed 493 amino acid protein that belongs to the TRIM/RBCC (Ring finger, B box, coiled-coil) family. TRIM35 contains a B box-type zinc finger, a coiled-coil domain, a SPRY domain and a RING-type zinc finger; a motif that has zinc-chelating activity and is involved in mediating protein-protein and protein-DNA interactions. Localizing to cytoplasmic granules and punctate nuclear bodies, TRIM35 is believed to play a role in the cell death mechanism. The forced expression of TRIM35 in HeLa cells results in the inhibition of tumorigenicity, cell growth and clonogenicity. In addition, the gene encoding TRIM35 localizes to a region of chromosome 8p21.2 that has been implicated in a number of leukemias and solid tumors. This suggests that TRIM35 may function as a tumor suppressor.

REFERENCES

1. Reymond, A., et al. 2001. The tripartite motif family identifies cell compartments. EMBO J. 20: 2140-2151.
2. Kimura, F., et al. 2003. Cloning and characterization of a novel RING-B-box-coiled-coil protein with apoptotic function. J. Biol. Chem. 278: 25046-25054.
3. Lalonde, J.P., et al. 2004. HLS5, a novel RBCC (Ring finger, B box, coiled-coil) family member isolated from a hemopoietic lineage switch, is a candidate tumor suppressor. J. Biol. Chem. 279: 8181-8189.
4. Kitamura, K., et al. 2005. The RING-finger protein haprin: domains and function in the acrosome reaction. Curr. Protein Pept. Sci. 6: 567-574.
5. Short, K.M. and Cox, T.C. 2006. Subclassification of the RBCC/TRIM superfamily reveals a novel motif necessary for microtubule binding. J. Biol. Chem. 281: 8970-8980.
6. Bouyain, S. and Leahy, D.J. 2007. Structure-based mutagenesis of the substrate-recognition domain of Nrdp1/FLRF identifies the binding site for the receptor tyrosine kinase ErbB3. Protein Sci. 16: 654-661.
7. Tao, H., et al. 2008. Structure of the MID1 tandem B-boxes reveals an interaction reminiscent of intermolecular ring heterodimers. Biochemistry 47: 2450-2457.

CHROMOSOMAL LOCATION

Genetic locus: TRIM35 (human) mapping to 8p21.2.

PRODUCT

TRIM35 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TRIM35 shRNA Plasmid (h): sc-76748-SH and TRIM35 shRNA (h) Lentiviral Particles: sc-76748-V as alternate gene silencing products.

For independent verification of TRIM35 (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-76748A, sc-76748B and sc-76748C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

TRIM35 siRNA (h) is recommended for the inhibition of TRIM35 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 µM in 66 µl. 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

TRIM35 (31-M): sc-100880 is recommended as a control antibody for monitoring of TRIM35 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 TRIM35 gene expression knockdown using RT-PCR Primer: TRIM35 (h)-PR: sc-76748-PR (20 µl). 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.