SANTA CRUZ BIOTECHNOLOGY, INC.

TRC8 siRNA (m): sc-63156

BACKGROUND

TRC8 (translocation in renal carcinoma on chromosome 8), also known as RNF139 (RING-finger protein 139), RCA1 or HRCA1 (hereditary renal cancer associated 1), is a multi-pass membrane protein that is predominantly expressed in testis, adrenal gland and placenta, and is expressed at lower levels in liver, skeletal muscle, pancreas, kidney, brain, heart and lung. Localizing to the endoplasmic reticulum (ER), TRC8 contains ten transmembrane segments, a sterol-sensing domain and one RING-type zinc finger, and may function as a ubiquitin ligase and signaling receptor. TRC8 physically interacts with VHL (von Hippel-Lindau disease tumor suppressor), and the inhibition of either of these proteins leads to the same ventral midline defect. Disruption of the TRC8 gene, caused by the 3;8 chromosomal translocation, is associated with hereditary renal cell carcinoma (RCC), suggesting that TRC8 is a potential tumor suppressor for RCC. Further supporting its role as a tumor suppressor, TRC8 mediates the induction of G_2/M phase arrest, increased apoptosis and decreased DNA synthesis.

REFERENCES

1. Boldog, F.L., et al. 1993. Positional cloning of the hereditary renal carcinoma 3;8 chromosome translocation breakpoint. Proc. Natl. Acad. Sci. USA 90: 8509-8513.
2. Gemmill, R.M., et al. 1998. The hereditary renal cell carcinoma 3;8 translocation fuses FHIT to a patched-related gene, TRC8. Proc. Natl. Acad. Sci. USA 95: 9572-9577.
3. Lorick, K.L., et al. 1999. RING fingers mediate ubiquitin-conjugating enzyme (E2)-dependent ubiquitination. Proc. Natl. Acad. Sci. USA 96: 11364-11369.
4. Gemmill, R.M., et al. 2002. The TRC8 hereditary kidney cancer gene suppresses growth and functions with VHL in a common pathway. Oncogene 21: 3507-3516.

CHROMOSOMAL LOCATION

Genetic locus: Rnf139 (mouse) mapping to 15 D1.

PRODUCT

TRC8 siRNA (m) 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 TRC8 shRNA Plasmid (m): sc-63156-SH and TRC8 shRNA (m) Lentiviral Particles: sc-63156-V as alternate gene silencing products.

For independent verification of TRC8 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-63156A, sc-63156B and sc-63156C.

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.

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

TRC8 siRNA (m) is recommended for the inhibition of TRC8 expression in mouse 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 60 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

TRC8 (H-9): sc-390347 is recommended as a control antibody for monitoring of TRC8 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 TRC8 gene expression knockdown using RT-PCR Primer: TRC8 (m)-PR: sc-63156-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.