

SUGT1 siRNA (m): sc-153916

BACKGROUND

SUGT1 (suppressor of G₂ allele of Skp1 p19 homolog, *S. cerevisiae*), also known as SGT1, is a homolog of the yeast protein Sgt1, a regulator of the cell cycle that is essential for G₁/S and G₂/M transitions. SUGT1 is a highly soluble protein and shares 26% overall amino acid identity and 30% overall similarity with its yeast counterpart. Localizing to the nucleus and cytoplasm, SUGT1 contains a CS domain, an SGS domain, a p23 domain and three tetratricopeptide repeats (TPR). The function of SUGT1 is conserved across eukaryotes. SUGT1 associates with Skp1 p19 and CUL-1, subunits of the SCF (Skp1-Cullin-F-box) ubiquitin ligase complex, and is believed to play a role in protein degradation. In addition, SUGT1 is required for the assembly of kinetochores and functions as a co-chaperone for HSP 90. An additional isoform, SUGT1B (also known as SGT1B), exists for SUGT1 due to alternative splicing events.

REFERENCES

1. Kitagawa, K., Skowrya, D., Elledge, S.J., Harper, J.W. and Hieter, P. 1999. SGT1 encodes an essential component of the yeast kinetochore assembly pathway and a novel subunit of the SCF ubiquitin ligase complex. *Mol. Cell* 4: 21-33.
2. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 604098. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Niikura, Y. and Kitagawa, K. 2003. Identification of a novel splice variant: human SGT1B (SUGT1B). *DNA Seq.* 14: 436-441.
4. Steensgaard, P., Garrè, M., Muradore, I., Transidico, P., Nigg, E.A., Kitagawa, K., Earnshaw, W.C., Faretta, M. and Musacchio, A. 2004. SGT1 is required for human kinetochore assembly. *EMBO Rep.* 5: 626-631.
5. Zou, X., Ji, C., Wang, L., Wu, M., Zheng, H., Xu, J., Jin, F., Gu, S., Ying, K., Xie, Y. and Mao, Y. 2004. Molecular cloning and characterization of SGT1.2, a novel splice variant of *Homo sapiens* SGT1. *DNA Seq.* 15: 140-143.
6. So, T., Takenoyama, M., Mizukami, M., Ichiki, Y., Sugaya, M., Hanagiri, T., Sugio, K. and Yasumoto, K. 2005. Haplotype loss of HLA class I antigen as an escape mechanism from immune attack in lung cancer. *Cancer Res.* 65: 5945-5952.

CHROMOSOMAL LOCATION

Genetic locus: Sugt1 (mouse) mapping to 14 D3.

PRODUCT

SUGT1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SUGT1 shRNA Plasmid (m): sc-153916-SH and SUGT1 shRNA (m) Lentiviral Particles: sc-153916-V as alternate gene silencing products.

For independent verification of SUGT1 (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-153916A, sc-153916B and sc-153916C.

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

SUGT1 siRNA (m) is recommended for the inhibition of SUGT1 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 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

SUGT1 (B-10): sc-398625 is recommended as a control antibody for monitoring of SUGT1 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 SUGT1 gene expression knockdown using RT-PCR Primer: SUGT1 (m)-PR: sc-153916-PR (20 μ l, 517 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.