

DEPTOR siRNA (h): sc-77660

BACKGROUND

DEPTOR (DEP domain containing MTOR-interacting protein), also known as DEP6 or DEPDC6 (DEP domain-containing protein 6), is a 409 amino acid protein that negatively regulates mTORC1 and mTORC2 pathways. DEPTOR interacts with FRAP via its PDZ domain, and undergoes post-translational phosphorylation. Containing two DEP domains and one PDZ (DHR) domain, DEPTOR is encoded by a gene that maps to human chromosome 8q24.12. Chromosome 8 consists of nearly 146 million base pairs, encodes over 800 genes and is associated with a variety of diseases and malignancies. Schizophrenia, bipolar disorder, trisomy 8, Pfeiffer syndrome, congenital hypothyroidism, Waardenburg syndrome and some leukemias and lymphomas are thought to occur as a result of defects in specific genes that map to chromosome 8.

REFERENCES

1. Kashino, G., et al. 2001. Preferential expression of an intact WRN gene in Werner syndrome cell lines in which a normal chromosome 8 has been introduced. *Biochem. Biophys. Res. Commun.* 289: 111-115.
2. Selicorni, A., et al. 2002. Cytogenetic mapping of a novel locus for type II Waardenburg syndrome. *Hum. Genet.* 110: 64-67.
3. McQueen, M.B., et al. 2005. Combined analysis from eleven linkage studies of bipolar disorder provides strong evidence of susceptibility loci on chromosomes 6q and 8q. *Am. J. Hum. Genet.* 77: 582-595.
4. Mossafa, H., et al. 2006. Non-Hodgkin's lymphomas with Burkitt-like cells are associated with c-Myc amplification and poor prognosis. *Leuk. Lymphoma* 47: 1885-1893.

CHROMOSOMAL LOCATION

Genetic locus: DEPTOR (human) mapping to 8q24.12.

PRODUCT

DEPTOR 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 DEPTOR shRNA Plasmid (h): sc-77660-SH and DEPTOR shRNA (h) Lentiviral Particles: sc-77660-V as alternate gene silencing products.

For independent verification of DEPTOR (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-77660A, sc-77660B and sc-77660C.

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

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

DEPTOR (A-3): sc-398169 is recommended as a control antibody for monitoring of DEPTOR 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 DEPTOR gene expression knockdown using RT-PCR Primer: DEPTOR (h)-PR: sc-77660-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Tan, M., et al. 2016. Depletion of SAG/RBX2 E3 ubiquitin ligase suppresses prostate tumorigenesis via inactivation of the PI3K/Akt/mTOR axis. *Mol. Cancer* 15: 81.

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.