SANTA CRUZ BIOTECHNOLOGY, INC.

CUL-7 siRNA (m): sc-60472

BACKGROUND

Cullin proteins comprise a distinct family of mediators that participate in the selective targeting of proteins for ubiquitin (Ub)-mediated proteolysis. CUL-7 mediates the third step of ubiquitin conjugation as part of an SCF-like complex consisting of CUL-7, Rbx1, Skp1, FBXW8 and GLMN isoform 1, which interacts with a complex of Skp1 and FBXW8, but not with Skp1 alone. This complex is thought to play a role in the degradation of proteins involved in proliferation and/or differentiation. CUL-7 is highly expressed in fetal kidney and adult skeletal muscle in addition to abundant expression in fetal brain and adult pancreas, kidney, placenta and heart. It is also detected in trophoblasts, lymphoblasts, osteoblasts, chondrocytes and skin fibroblasts. Defects in the gene encoding CUL-7 result in 3-M syndrome, an autosomal recessive disorder characterized by severe pre- and postnatal growth retardation, facial dysmorphism, large head circumference and normal intelligence and endocrine function as well as skeletal changes including long slender tubular bones and tall vertebral bodies.

REFERENCES

1. Kipreos, E.T., et al. 1996. CUL-1 is required for cell cycle exit in *C. elegans* and identifies a novel gene family. Cell 85: 829-839.
2. Dias, D.C., et al. 2002. CUL-7: A DOC domain-containing cullin selectively binds Skp1.FBX29 to form an SCF-like complex. Proc. Natl. Acad. Sci. USA 99: 16601-16606.
3. Arai, T., et al. 2003. Targeted disruption of p185/CUL-7 gene results in abnormal vascular morphogenesis. Proc. Natl. Acad. Sci. USA 100: 9855-9860.
4. Huber, C., et al. 2005. Identification of mutations in CUL-7 in 3-M syndrome. Nat. Genet. 37: 1119-1124.
5. Skaar, J.R., et al. 2005. Dimerization of CUL-7 and PARC is not required for all CUL-7 functions and mouse development. Mol. Cell. Biol. 25: 5579-5589.
6. Andrews, P., et al. 2006. Cytoplasmic localized ubiquitin ligase growth by antagonizing p53 function. Oncogene 25: 4534-4548.
7. Kasper, J.S., et al. 2006. A novel p53-binding domain in CUL-7. Biochem. Biophys. Res. Commun. 348: 132-138.

CHROMOSOMAL LOCATION

Genetic locus: Cul7 (mouse) mapping to 17 C.

PRODUCT

CUL-7 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 CUL-7 shRNA Plasmid (m): sc-60472-SH and CUL-7 shRNA (m) Lentiviral Particles: sc-60472-V as alternate gene silencing products.

For independent verification of CUL-7 (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-60472A, sc-60472B and sc-60472C.

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

CUL-7 siRNA (m) is recommended for the inhibition of CUL-7 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 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

CUL-7 (H-8): sc-514970 is recommended as a control antibody for monitoring of CUL-7 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 CUL-7 gene expression knockdown using RT-PCR Primer: CUL-7 (m)-PR: sc-60472-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.