

DBR1 siRNA (h): sc-77921

BACKGROUND

DBR1 (debranching enzyme homolog 1) is a 544 amino acid protein that localizes to the nucleus and belongs to the lariat debranching enzyme family. Functioning at an optimal pH of 7.0, DBR1 uses divalent metal cations to catalyze the cleavage of the 2'-5' phosphodiester linkage at the branch point of lariat intron pre-mRNAs, thereby converting the lariat structures to linear molecules that are subject to degradation. Via its catalytic activity, DBR1 facilitates ribonucleotide turnover and is thought to participate in retroviral (specifically HIV-1) replication. Human DBR1 shares 79% homology with its mouse counterpart, suggesting a conserved role between species. DBR1 is expressed as two alternatively spliced isoforms that are encoded by a gene which maps to human chromosome 3q22.3.

REFERENCES

1. Arenas, J. and Hurwitz, J. 1987. Purification of a RNA debranching activity from HeLa cells. *J. Biol. Chem.* 262: 4274-4279.
2. Chapman, K.B. and Boeke, J.D. 1991. Isolation and characterization of the gene encoding yeast debranching enzyme. *Cell* 65: 483-492.
3. Kim, J.W., et al. 2000. Human RNA lariat debranching enzyme cDNA complements the phenotypes of *Saccharomyces cerevisiae* DBR1 and *Schizosaccharomyces pombe* DBR1 mutants. *Nucleic Acids Res.* 28: 3666-3673.
4. Martin, A., et al. 2002. Prp43 is an essential RNA-dependent ATPase required for release of lariat-intron from the spliceosome. *J. Biol. Chem.* 277: 17743-17750.

CHROMOSOMAL LOCATION

Genetic locus: DBR1 (human) mapping to 3q22.3.

PRODUCT

DBR1 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 DBR1 shRNA Plasmid (h): sc-77921-SH and DBR1 shRNA (h) Lentiviral Particles: sc-77921-V as alternate gene silencing products.

For independent verification of DBR1 (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-77921A, sc-77921B and sc-77921C.

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

DBR1 siRNA (h) is recommended for the inhibition of DBR1 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

DBR1 (3A7): sc-517060 is recommended as a control antibody for monitoring of DBR1 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 DBR1 gene expression knockdown using RT-PCR Primer: DBR1 (h)-PR: sc-77921-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. Choi, S., et al. 2022. Non-canonical splice junction processing increases the diversity of RBFOX2 splicing isoforms. *Int. J. Biochem. Cell Biol.* 144: 106172.

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.