

TRIAD1 siRNA (m): sc-63160

BACKGROUND

TRIAD1, also known as ARIH2 (ariadne homolog 2) or ARI2, is a 493 amino acid protein that contains one IBR-type zinc finger and 2 RING-type zinc fingers and belongs to the ariadne subfamily of RBR proteins. Localized to the nucleus, TRIAD1 interacts with UBE2L3 and is thought to act as an E3 ubiquitin-protein ligase, functioning to accept ubiquitin from E2 ubiquitin-conjugating enzymes and transfer the acquired ubiquitin residue to target substrates. TRIAD1 is subject to post-translational DNA damage-dependent phosphorylation, probably by ATM or ATR. The gene encoding TRIAD1 maps to human chromosome 3, which houses over 1,100 genes, including a chemokine receptor (CKR) gene cluster and a variety of human cancer-related gene loci.

REFERENCES

1. van der Reijden, B.A., et al. 1999. TRIADs: a new class of proteins with a novel cysteine-rich signature. *Protein Sci.* 8: 1557-1561.
2. Aguilar, M., et al. 2000. Ariadne-1: a vital *Drosophila* gene is required in development and defines a new conserved family of RING-finger proteins. *Genetics* 155: 1231-1244.
3. Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 605615. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Beitel, L.K., et al. 2002. Cloning and characterization of an androgen receptor N-terminal-interacting protein with ubiquitin-protein ligase activity. *J. Mol. Endocrinol.* 29: 41-60.
5. Marteijn, J.A., et al. 2005. The E3 ubiquitin-protein ligase TRIAD1 inhibits clonogenic growth of primary myeloid progenitor cells. *Blood* 106: 4114-4123.
6. Marteijn, J.A., et al. 2007. Gfi1 ubiquitination and proteasomal degradation is inhibited by the ubiquitin ligase TRIAD1. *Blood* 110: 3128-3135.
7. Matsuoka, S., et al. 2007. Atm and ATR substrate analysis reveals extensive protein networks responsive to DNA damage. *Science* 316: 1160-1166.

CHROMOSOMAL LOCATION

Genetic locus: Arih2 (mouse) mapping to 9 F2.

PRODUCT

TRIAD1 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 TRIAD1 shRNA Plasmid (m): sc-63160-SH and TRIAD1 shRNA (m) Lentiviral Particles: sc-63160-V as alternate gene silencing products.

For independent verification of TRIAD1 (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-63160A, sc-63160B and sc-63160C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

TRIAD1 siRNA (m) is recommended for the inhibition of TRIAD1 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

TRIAD1 (C-7): sc-390682 is recommended as a control antibody for monitoring of TRIAD1 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 TRIAD1 gene expression knockdown using RT-PCR Primer: TRIAD1 (m)-PR: sc-63160-PR (20 μ l, 515 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.