

SANTA CRUZ BIOTECHNOLOGY, INC.

ATR siRNA (m): sc-29764



The Power to Ouestion

BACKGROUND

Members of the PIK (phosphatidylinositol kinase)-related kinase family are high molecular weight kinases involved in cell cycle progression, DNA recombination and detection of DNA damage. One member of the Pl3/Pl4-kinase family is ATR (ataxia-telangiectasia- and Rad3-related), also known as FRP1 (for FRAP-related protein 1). ATR is most closely related to ATM, a protein kinase encoded by the gene mutated in ataxia telangiectasia. ATR is also closely related to three of the family members involved in checkpoint function: Mei-41 (*Drosophila*), Mec1p (*S. cerevisiae*) and Rad3 (*Schizosaccharomyces pombe*), and as such may be the functional human counterpart of these proteins. This kinase has been shown to phosphorylate checkpoint kinase CHK1, checkpoint proteins Rad17 and Rad9, as well as tumor suppressor protein BRCA1. In addition, ATR is essential for early embryonic development. The human ATR gene maps to chromosome 3q22-q24, encodes a 300 kDa protein and localizes to intranuclear foci after DNA damage or inhibition of replication.

REFERENCES

- Cimprich, K., et al. 1996. cDNA cloning and gene mapping of a candidate human cell cycle checkpoint protein. Proc. Natl. Acad. Sci. USA 93: 2850- 2855.
- Bao, S., et al. 2001. ATR/ATM-mediated phosphorylation of human Rad17 is required for genotoxic stress responses. Nature 411: 969-974.
- 3. Online Mendelian Inheritance in Man, OMIM™. 2004. Johns Hopkins University, Baltimore, MD. MIM Number: 601215. World Wide Web URL: http://www.ncbi.nlm. nih.gov/omim/.
- 4. SWISS-PROT/TrEMBL (NP_001175). World Wide Web URL: http://www.expasy.ch/sprot/sprot-top.html.

CHROMOSOMAL LOCATION

Genetic locus: ATR (human) mapping to 3q22-q24; Atr (mouse) mapping to 9 E4.

PRODUCT

ATR siRNA (m) is a pool of 3 target-specific 20-25 nt siRNAs designed to knock down gene expression. Each vial contains 3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections.

For independent verification of ATR (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-29764A, sc-29764B and sc-29764C.

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

ATR siRNA (m) is recommended for the inhibition of ATR expression in mouse cells.

ATR (N-19): sc-1887 is recommended as a control antibody for Western Blotting (starting dilution 1:100, dilution range 1:100-1:1,000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) protein detection using the recommended secondary reagents listed below.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

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 10µM in 60 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. Semi-quantitative RT-PCR may be performed using RT-PCR Primer: ATR (m)-PR: sc-29764-PR (20 µl).

RESEARCH USE

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

PROTOCOLS

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