SANTA CRUZ BIOTECHNOLOGY, INC.

ATR siRNA (h): sc-29763

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 PI 3-/PI 4-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 protein encoded by the human ATR gene localizes to intranuclear foci after DNA damage or inhibition of replication.

REFERENCES

1. 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.
2. Keegan, K., et al. 1996. The ATR and ATM protein kinases associate with different sites along meiotically pairing chromosomes. Genes Dev. 10: 2423-2437.

CHROMOSOMAL LOCATION

Genetic locus: ATR (human) mapping to 3q23.

PRODUCT

ATR 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ATR shRNA Plasmid (h): sc-29763-SH and ATR shRNA (h) Lentiviral Particles: sc-29763-V as alternate gene silencing products.

For independent verification of ATR (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-29763A, sc-29763B and sc-29763C.

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

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

ATR (C-1): sc-515173 is recommended as a control antibody for monitoring of ATR 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ATR gene expression knockdown using RT-PCR Primer: ATR (h)-PR: sc-29763-PR (20 µl, 315 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Auclair, Y., et al. 2008. ATR kinase is required for global genomic nucleotide excision repair exclusively during S phase in human cells. Proc. Natl. Acad. Sci. USA 105: 17896-17901.
2. Fan, S., et al. 2009. Low concentrations of diindolylmethane, a metabolite of indole-3-carbinol, protect against oxidative stress in a BRCA1-dependent manner. Cancer Res. 69: 6083-6091.
3. Hahm, S.H., et al. 2012. Human MutY homolog induces apoptosis in etoposide-treated HEK293 cells. Oncol. Lett. 4: 1203-1208.
4. Yuan, F., et al. 2013. Overexpressed DNA polymerase iota regulated by JNK/c-Jun contributes to hypermutagenesis in bladder cancer. PLoS ONE 8: e69317.
5. Brown, A.D., et al. 2014. ATR suppresses endogenous DNA damage and allows completion of homologous recombination repair. PLoS ONE 9: e91222.
6. Khanal, S. and Galloway, D.A. 2019. High-risk human papillomavirus oncogenes disrupt the Fanconi anemia DNA repair pathway by impairing localization and de-ubiquitination of FANCD2. PLoS Pathog. 15: e1007442.
7. Hiregange, D., et al. 2020. ATR signalling mediates the prosurvival function of phospho-NPM against PIDDosome mediated cell death. Cell. Signal. 71: 109602.
8. Yu, X., et al. 2020. Ubiquitination of the DNA-damage checkpoint kinase CHK1 by TRAF4 is required for CHK1 activation. J. Hematol. Oncol. 13: 40.

RESEARCH USE

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