SANTA CRUZ BIOTECHNOLOGY, INC.

PKR siRNA (h): sc-36263

BACKGROUND

An interferon-inducible, RNA-dependent protein serine/threonine kinase (PKR) has been described. PKR in earlier literature is variously known as DAI, dsJ, PI kinase, p65, p67 or TIK for the mouse kinase; and p68 or p69 for the human kinase. The PKR kinase substrate is the a subunit of protein synthesis initiation factor eIF-2. Phosphorylation of eIF-2α on Serine-51 results in inhibition of translation. Molecular cDNA clones have been isolated from both human and mouse cells. The serine/threonine kinase catalytic domains map to the carboxy terminal half of the protein while the RNA-binding domains are located in the amino terminal region. Three kinds of regulation of PKR enzymatic activity have been described. These include transcriptional regulation in response to interferon, an autoregulatory mechanism controlling PKR expression at the level of translation and post-translational regulation by RNA mediated autophosphorylation.

REFERENCES

1. Hershey, J.W. 1989. Protein phosphorylation controls translation rates. J. Biol. Chem. 264: 20823-20826.
2. Meurs, E., et al. 1990. Molecular cloning and characterization of the human double-stranded RNA-activated protein kinase induced by interferon. Cell 62: 379-390.

CHROMOSOMAL LOCATION

Genetic locus: EIF2AK2 (human) mapping to 2p22.2.

PRODUCT

PKR 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 PKR shRNA Plasmid (h): sc-36263-SH and PKR shRNA (h) Lentiviral Particles: sc-36263-V as alternate gene silencing products.

For independent verification of PKR (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-36263A, sc-36263B and sc-36263C.

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

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

PKR (B-10): sc-6282 is recommended as a control antibody for monitoring of PKR 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 PKR gene expression knockdown using RT-PCR Primer: PKR (h)-PR: sc-36263-PR (20 µl, 570 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Sarkar, D., et al. 2007. Activation of double-stranded RNA dependent protein kinase, a new pathway by which human polynucleotide phosphorylase (hPNPase(old-35)) induces apoptosis. Cancer Res. 67: 7948-7953.
2. Sanghvi, V.R. and Steel, L.F. 2011. The cellular TAR RNA binding protein, TRBP, promotes HIV-1 replication primarily by inhibiting the activation of double-stranded RNA-dependent kinase PKR. J. Virol. 85: 12614-12621.
3. Dai, R., et al. 2012. Activation of PKR/eIF2α signaling cascade is associated with dihydrotestosterone-induced cell cycle arrest and apoptosis in human liver cells. J. Cell. Biochem. 113: 1800-1808.
4. Zhang, L., et al. 2013. The modulation of hepatitis C virus 1a replication by PKR is dependent on NFκB mediated interferon beta response in Huh7.5.1 cells. Virology 438: 28-36.
5. Wei, C., et al. 2015. Involvement of general control nonderepressible kinase 2 in cancer cell apoptosis by posttranslational mechanisms. Mol. Biol. Cell 26: 1044-1057.
6. Fu, X., et al. 2016. Malonate induces the assembly of cytoplasmic stress granules. FEBS Lett. 590: 22-33.
7. Tang, Y., et al. 2017. Polycystin-1 inhibits eIF2α phosphorylation and cell apoptosis through a PKR-eIF2α pathway. Sci. Rep. 7: 11493.
8. Christ, W., et al. 2020. Puumala and Andes hantaviruses cause transient PKR-dependent formation of stress granules. J. Virol. 94: e01168-19.

RESEARCH USE

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