

PKR siRNA (m): sc-36264

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 α subunit of protein synthesis initiation factor eIF-2. Phosphorylation of eIF-2 α 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

- Hershey, J.W. 1989. Protein phosphorylation controls translation rates. *J. Biol. Chem.* 264: 20823-20826.
- 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 (mouse) mapping to 17 E3.

PRODUCT

PKR 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 PKR shRNA Plasmid (m): sc-36264-SH and PKR shRNA (m) Lentiviral Particles: sc-36264-V as alternate gene silencing products.

For independent verification of PKR (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-36264A, sc-36264B and sc-36264C.

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

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

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 (m)-PR: sc-36264-PR (20 μ l, 503 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Pataer, A., et al. 2009. Inhibition of RNA-dependent protein kinase (PKR) leads to cancer cell death and increases chemosensitivity. *Cancer Biol. Ther.* 8: 245-252.
- Hage Hassan, R., et al. 2016. Sustained action of ceramide on the Insulin signaling pathway in muscle cells: implication of the double-stranded RNA-activated protein kinase. *J. Biol. Chem.* 291: 3019-3029.
- Zhang, L., et al. 2016. Interferon β (IFN- β) production during the double-stranded RNA (dsRNA) response in hepatocytes involves coordinated and feedforward signaling through Toll-like receptor 3 (TLR3), RNA-dependent protein kinase (PKR), inducible nitric oxide synthase (iNOS), and Src protein. *J. Biol. Chem.* 291: 15093-15107.
- Fan, X., et al. 2017. Foot-and-mouth disease virus infection suppresses autophagy and NF κ B antiviral responses via degradation of ATG5-ATG12 by 3C^{pro}. *Cell Death Dis.* 8: e2561.
- Teramachi, J., et al. 2017. PKR regulates LPS-induced osteoclast formation and bone destruction *in vitro* and *in vivo*. *Oral Dis.* 23: 181-188.
- Lan, T., et al. 2020. Peroxynitrite/PKR axis modulates the NLRP3 inflammasome of cardiac fibroblasts. *Front. Immunol.* 11: 558712.
- Gurung, C., et al. 2021. Dicer represses the interferon response and the double-stranded RNA-activated protein kinase pathway in mouse embryonic stem cells. *J. Biol. Chem.* 296: 100264.

RESEARCH USE

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