SANTA CRUZ BIOTECHNOLOGY, INC.

DAP-4 siRNA (m): sc-77094

BACKGROUND

In contrast to growth factors which promote cell proliferation, FAS ligand (FAS-L) and the tumor necrosis factors (TNFs) rapidly induce apoptosis. Cellular response to FAS-L and TNF is mediated by structurally related receptors containing a conserved cytoplasmic region called the death domain. DAP-4 (death-associated protein 4), also known as PRKRIR (protein-kinase, interferon-inducible double stranded RNA dependent inhibitor, repressor of (P58 repressor)), p58IPK-interacting protein or THAP0 (THAP domain-containing protein 0), is a 761 amino acid protein that functions as an upstream repressor of interferon-induced PKR. Containing one THAP-type zinc finger and existing as two alternatively spliced isoforms, DAP-4 inhibits the PKR-inhibitory ability of DnaJC3 (also known as P58IPK), leading to suppression of cell growth and restoration of kinase activity.

REFERENCES

1. Mellor, H., et al. 1991. A synthetic peptide substrate for initiation factor-2 kinases. Biochem. Biophys. Res. Commun. 178: 430-437.
2. Polyak, S.J., et al. 1996. The P58 cellular inhibitor complexes with the interferon-induced, double-stranded RNA-dependent protein kinase, PKR, to regulate its autophosphorylation and activity. J. Biol. Chem. 271: 1702-1707.
3. Shi, Y., et al. 1998. Identification and characterization of pancreatic eukaryotic initiation factor 2 α-subunit kinase, PEK, involved in translational control. Mol. Cell. Biol. 18: 7499-7509.
4. Gale, M., et al. 1998. Regulation of interferon-induced protein kinase PKR: modulation of P58IPK inhibitory function by a novel protein, P52rIPK. Mol. Cell. Biol. 18: 859-871.
5. Lu, J., et al. 1999. The interferon-induced double-stranded RNA-activated protein kinase PKR will phosphorylate serine, threonine, or tyrosine at residue 51 in eukaryotic initiation factor 2α. J. Biol. Chem. 274: 32198-32203.

CHROMOSOMAL LOCATION

Genetic locus: Prkrir (mouse) mapping to 7 E2.

PRODUCT

DAP-4 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see DAP-4 shRNA Plasmid (m): sc-77094-SH and DAP-4 shRNA (m) Lentiviral Particles: sc-77094-V as alternate gene silencing products.

For independent verification of DAP-4 (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-77094A, sc-77094B and sc-77094C.

PROTOCOLS

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

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

DAP-4 siRNA (m) is recommended for the inhibition of DAP-4 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor DAP-4 gene expression knockdown using RT-PCR Primer: DAP-4 (m)-PR: sc-77094-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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