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DJ-1 siRNA (m2): sc-270201

BACKGROUND

The DJ-1 gene encodes a highly-conserved protein which is implicated in a number of cell processes. DJ-1 was first identified as a novel oncogene that transformed mouse NIH/3T3 cells in cooperation with activated Ras. Additionally, DJ-1 was cloned in rat as SP22 or CAP-1, and found to be an infertility-related sperm protein whose expression is reduced in sperm treated with toxicants. DJ-1 also positively regulates the androgen receptor (AR) by forming a complex with PIAS α , a negative regulator of AR. The gene encoding human DJ-1 maps to chromosome 1p36.23, a region identified as a hot spot of chromosome abnormalities in several tumor cells. Subsequently, mutations in the DJ-1 gene have been implicated in Parkinson's disease, and loss of DJ-1 function leads to neurodegeneration. DJ-1 is a ubiquitously expressed protein that is induced in response to growth stimuli and translocates from the cytoplasm to the nucleus during the S phase of the cell cycle.

REFERENCES

1. Nagakubo, D., et al. 1997. DJ-1, a novel oncogene which transforms mouse NIH/3T3 cells in cooperation with Ras. *Biochem. Biophys. Res. Commun.* 231: 509-513.
2. Taira, T., et al. 2001. Molecular cloning of human and mouse DJ-1 genes and identification of Sp1-dependent activation of the human DJ-1 promoter. *Gene* 263: 285-292.
3. Takahashi, K., et al. 2001. DJ-1 positively regulates the androgen receptor by impairing the binding of PIAS α to the receptor. *J. Biol. Chem.* 276: 37556-37563.
4. Bonifati, V., et al. 2003. Mutations in the DJ-1 gene associated with autosomal recessive early-onset parkinsonism. *Science* 299: 256-259.
5. Niki, T., et al. 2003. DJBP: A novel DJ-1-binding protein, negatively regulates the androgen receptor by recruiting histone deacetylase complex, and DJ-1 antagonizes this inhibition by abrogation of this complex. *Mol. Cancer Res.* 1: 247-261.
6. Cookson, M.R. 2003. Pathways to parkinsonism. *Neuron* 37: 7-10.

CHROMOSOMAL LOCATION

Genetic locus: Park7 (mouse) mapping to 4 E2.

PRODUCT

DJ-1 siRNA (m2) is a pool of 2 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 DJ-1 shRNA Plasmid (m2): sc-270201-SH and DJ-1 shRNA (m2) Lentiviral Particles: sc-270201-V as alternate gene silencing products.

For independent verification of DJ-1 (m2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270201A and sc-270201B.

RESEARCH USE

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

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

DJ-1 siRNA (m2) is recommended for the inhibition of DJ-1 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

DJ-1 (D-4): sc-55572 is recommended as a control antibody for monitoring of DJ-1 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).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor DJ-1 gene expression knockdown using RT-PCR Primer: DJ-1 (m2)-PR: sc-270201-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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