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PINK1 siRNA (m): sc-44599

BACKGROUND

A member of the serine/threonine protein kinase family, PTEN induced putative kinase 1 (PINK1) is a tumor suppressor. PINK1 is primarily located in mitochondria, and is ubiquitously expressed in testis, skeletal muscle, and heart tissue. It can also be detected at lower levels in pancreas, ovary, brain, placenta, kidney, liver, prostate and small intestine. During cellular stress PINK1 protects against mitochondrial dysfunction by inducing phosphorylation mitochondrial proteins. PINK1 mutations may give rise to different autophosphorylation activity. Mutations in the PINK1 gene (PARK6) are associated with early onset Parkinson's disease, a recessive neurodegenerative disorder characterized by resting tremor, muscular rigidity, bradykinesia and postural instability. Parkinson's disease generally involves the presence of intraneuronal accumulations of aggregated proteins (Lewy bodies) in brain neurons.

CHROMOSOMAL LOCATION

Genetic locus: Pink1 (mouse) mapping to 4 D3.

PRODUCT

PINK1 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 PINK1 shRNA Plasmid (m): sc-44599-SH and PINK1 shRNA (m) Lentiviral Particles: sc-44599-V as alternate gene silencing products.

For independent verification of PINK1 (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-44599A, sc-44599B and sc-44599C.

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

PINK1 siRNA (m) is recommended for the inhibition of PINK1 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

PINK1 (38CT20.8.5): sc-517353 is recommended as a control antibody for monitoring of PINK1 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 PINK1 gene expression knockdown using RT-PCR Primer: PINK1 (m)-PR: sc-44599-PR (20 μ l, 564 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Shen, M., et al. 2016. FSH protects mouse granulosa cells from oxidative damage by repressing mitophagy. *Sci. Rep.* 6: 38090.
- Thangaraj, A., et al. 2018. HIV-1 Tat-mediated microglial activation: role of mitochondrial dysfunction and defective mitophagy. *Autophagy* 14: 1596-1619.
- Chen, K., et al. 2018. Optineurin-mediated mitophagy protects renal tubular epithelial cells against accelerated senescence in diabetic nephropathy. *Cell Death Dis.* 9: 105.
- Thangaraj, A., et al. 2019. Mitigation of cocaine-mediated mitochondrial damage, defective mitophagy and microglial activation by superoxide dismutase mimetics. *Autophagy* 16: 289-312.
- Zhong, L., et al. 2020. Phosphorylation of cGAS by CDK1 impairs self-DNA sensing in mitosis. *Cell Discov.* 6: 26.
- Zhou, P., et al. 2019. Notoginsenoside R1 ameliorates diabetic retinopathy through PINK1-dependent activation of mitophagy. *Cells* 8: 213.
- Li, X., et al. 2020. Cyanidin-3-O-glucoside improves non-alcoholic fatty liver disease by promoting PINK1-mediated mitophagy in mice. *Br. J. Pharmacol.* E-published.

RESEARCH USE

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

PROTOCOLS

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