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PON1 siRNA (h): sc-44031

BACKGROUND

Paroxon is an organophosphorus anticholinesterase compound, used topically in the treatment of glaucoma. It is produced *in vivo* in mammals by microsomal oxidation of the insecticide parathion. Parathion is inert until transformed to paroxon. Paroxonase or PON is an arylesterase that is capable of hydrolyzing paroxon to produce p-nitrophenol. PONs are nonspecific and their classification is based not only on substrate specificity but also on tissue distribution, inhibition properties and physicochemical characteristics such as electrophoretic mobility and molecular weight. In contrast to PON1, which is expressed mainly in the liver, PON2 is expressed in a variety of mouse tissues, including the pancreas. PON3 is associated with the high density lipoprotein fraction of serum. The genes which encode PON1-3 are physically linked and map to human chromosome 7q21.3.

REFERENCES

1. Coates, P.M., et al. 1975. A preliminary genetic interpretation of the esterase isozymes of human tissues. *Ann. Hum. Genet.* 39: 1-20.
2. Humbert, R., et al. 1993. The molecular basis of the human serum paraoxonase activity polymorphism. *Nat. Genet.* 3: 73-76.
3. Primo-Parmo, S.L., et al. 1996. The human serum paraoxonase/arylesterase gene (PON1) is one member of a multigene family. *Genomics* 33: 498-507.
4. Mochizuki, H., et al. 1998. Human PON2 gene at 7q21.3: cloning, multiple mRNA forms, and missense polymorphisms in the coding sequence. *Gene* 213: 149-157.
5. Draganov, D.I., et al. 2000. Rabbit serum paraoxonase 3 (PON3) is a high density lipoprotein-associated lactonase and protects low density lipoprotein against oxidation. *J. Biol. Chem.* 275: 33435-33442.
6. LocusLink Report (LocusID: 168820). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: PON1 (human) mapping to 7q21.3.

PRODUCT

PON1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PON1 shRNA Plasmid (h): sc-44031-SH and PON1 shRNA (h) Lentiviral Particles: sc-44031-V as alternate gene silencing products.

For independent verification of PON1 (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-44031A, sc-44031B and sc-44031C.

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.

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

PON1 siRNA (h) is recommended for the inhibition of PON1 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 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

PON1 (17A12): sc-59646 is recommended as a control antibody for monitoring of PON1 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 PON1 gene expression knockdown using RT-PCR Primer: PON1 (h)-PR: sc-44031-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.