

OAT siRNA (h): sc-62709

BACKGROUND

OAT (ornithine aminotransferase (mitochondrial), ornithine-oxo-acid aminotransferase) is a 439 amino acid protein encoded by the human gene OAT. OAT belongs to the class III pyridoxal-phosphate-dependent aminotransferase family and is usually found as a homotetramer in the mitochondrion matrix. OAT catalyzes the major catalytic reaction for ornithine. Ornithinemia, presumably due to deficiency of ornithine ketoacid aminotransferase (OAT) has been found in patients with gyrate atrophy of the choroid and retina. The clinical history of gyrate atrophy is usually night blindness that begins in late childhood, accompanied by sharply demarcated circular areas of chorioretinal atrophy. During the second and third decades the areas of atrophy enlarge. The hepatic cleavage product, hepatic OAT, is formed by cleaving a 25 amino acid transit peptide from the N-terminus of the OAT precursor. The renal form is produced by cleaving a 35 amino acid transit peptide from the N-terminus.

REFERENCES

1. Ramesh, V., et al. 1991. Molecular pathology of gyrate atrophy of the choroid and retina due to ornithine aminotransferase deficiency. *Mol. Biol. Med.* 8: 81-93.
2. Michaud, J., et al. 1992. Strand-separating conformational polymorphism analysis: efficacy of detection of point mutations in the human ornithine δ -aminotransferase gene. *Genomics* 13: 389-394.
3. Shah, S.A., et al. 1997. Human ornithine aminotransferase complexed with L-canaline and gabaculine: structural basis for substrate recognition. *Structure* 5: 1067-1075.
4. Buard, J., et al. 2000. Somatic versus germline mutation processes at minisatellite CEB1 (D2S90) in humans and transgenic mice. *Genomics* 65: 95-103.
5. Cleary, M.A., et al. 2005. Ornithine aminotransferase deficiency: diagnostic difficulties in neonatal presentation. *J. Inher. Metab. Dis.* 28: 673-679.

CHROMOSOMAL LOCATION

Genetic locus: OAT (human) mapping to 10q26.13.

PRODUCT

OAT 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 OAT shRNA Plasmid (h): sc-62709-SH and OAT shRNA (h) Lentiviral Particles: sc-62709-V as alternate gene silencing products.

For independent verification of OAT (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-62709A, sc-62709B and sc-62709C.

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

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

OAT (A-12): sc-374243 is recommended as a control antibody for monitoring of OAT 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 OAT gene expression knockdown using RT-PCR Primer: OAT (h)-PR: sc-62709-PR (20 μ l, 578 bp). 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.