SANTA CRUZ BIOTECHNOLOGY, INC.

AOX1 siRNA (h): sc-94924

BACKGROUND

The formation of free radicals is an adverse consequence of metabolism. Free radicals endanger cells by causing oxidative damage to membranes and can lead to interruption of DNA sequences, thereby potentially resulting in carcinogenesis. As a member of the molybdo-flavoenzymes family of proteins, AOX1 (Aldehyde oxidase 1) is a 1,338 amino acid cytoplasmic protein that catalyzes the oxidation of a variety of aldehydes, leading to the production of hydrogen peroxide. Under certain conditions, AOX1 can catalyze the formation of the superoxide free radical. Defects in oxygen radical metabolism have been linked to the pathogenesis of amyotrophic lateral sclerosis (ALS), an autosomal dominant neurodegenerative disorder characterized by the death of motor neurons in the spinal cord, brain and brainstem. Significantly, AOX1 is highly expressed in the ventral horn of the spinal cord and the gene that encodes AOX1 is located in a chromosomal region that is frequently found to be implicated in ALS2. This evidence suggests that AOX1 is a candidate gene for ALS2.

REFERENCES

1. Wright, R.M., et al. 1993. cDNA cloning, characterization, and tissue-specific expression of human xanthine dehydrogenase/xanthine oxidase. Proc. Natl. Acad. Sci. USA 90: 10690-10694.
2. Berger, R., et al. 1995. Analysis of aldehyde oxidase and xanthine dehydrogenase/oxidase as possible candidate genes for autosomal recessive familial amyotrophic lateral sclerosis. Somat. Cell Mol. Genet. 21: 121-131.
3. Online Mendelian Inheritance in Man, OMIM™. 1998. Johns Hopkins University, Baltimore, MD. MIM Number: 602841. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Kurosaki, M., et al. 2004. The aldehyde oxidase gene cluster in mice and rats. Aldehyde oxidase homologue 3, a novel member of the molybdo-flavoenzyme family with selective expression in the olfactory mucosa. J. Biol. Chem. 279: 50482-50498.

CHROMOSOMAL LOCATION

Genetic locus: AOX1 (human) mapping to 2q33.1.

PRODUCT

AOX1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see AOX1 shRNA Plasmid (h): sc-94924-SH and AOX1 shRNA (h) Lentiviral Particles: sc-94924-V as alternate gene silencing products.

For independent verification of AOX1 (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-94924A, sc-94924B and sc-94924C.

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

AOX1 siRNA (h) is recommended for the inhibition of AOX1 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 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.

GENE EXPRESSION MONITORING

AOX1 (D-8): sc-365291 is recommended as a control antibody for monitoring of AOX1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 AOX1 gene expression knockdown using RT-PCR Primer: AOX1 (h)-PR: sc-94924-PR (20 µl, 571 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.