SANTA CRUZ BIOTECHNOLOGY, INC.

ARALAR siRNA (m): sc-141183

BACKGROUND

Calcium signaling in mitochondria is important in order for it to function in response to a variety of extracellular stimuli. Signaling begins with Ca^{2+} entry in mitochondria via the Ca^{2+} uniporter followed by Ca^{2+} activation of three dehydrogenases in the mitochondrial matrix. ARALAR, the neuronal Ca^{2+}-binding mitochondrial aspartate-glutamate carrier, has Ca^{2+} binding domains facing the extramitochondrial space and functions in the malate-aspartate NADH shuttle (MAS). ARALAR is encoded by the SLC25a12 gene and is expressed in brain and skeletal muscle. ARALAR is required for the synthesis of brain aspartate and N-acetylaspartatemay and plays a role in myelin formation. It is also essential for the transmission of small Ca^{2+} signals to mitochondria via an increase in mitochondrial NADH. In addition, ARALAR is implicated in conferring susceptibility to schizophrenia.

REFERENCES

1. Jalil, M.A., et al. 2005. Reduced N-acetylaspartate levels in mice lacking aralar, a brain- and muscle-type mitochondrial aspartate-glutamate carrier. J. Biol. Chem. 280: 31333-31339.
2. Pardo, B., et al. 2006. Essential role of aralar in the transduction of small Ca^{2+} signals to neuronal mitochondria. J. Biol. Chem. 281: 1039-1047.
3. Contreras, L., et al. 2007. Ca^{2+} Activation kinetics of the two aspartate-glutamate mitochondrial carriers, ARALAR and citrin: role in the heart malate-aspartate NADH shuttle. J. Biol. Chem. 282: 7098-7106.
4. Satrústegui, J., et al. 2007. Role of ARALAR, the mitochondrial transporter of aspartate-glutamate, in brain N-acetylaspartate formation and Ca^{2+} signaling in neuronal mitochondria. J. Neurosci. Res. 85: 3359-3366.
5. Satrústegui, J., et al. 2007. Mitochondrial transporters as novel targets for intracellular calcium signaling. Physiol. Rev. 87: 29-67.
6. Hong, C.J., et al. 2007. Association study of polymorphisms in the mitochondrial aspartate/glutamate carrier SLC25A12 (ARALAR) gene with schizophrenia. Prog. Neuropsychopharmacol. Biol. Psychiatry 31: 1510-1513.
7. Mármol, P., et al. 2009. Requirement for ARALAR and its Ca^{2+}-binding sites in Ca^{2+} signal transduction in mitochondria from INS-1 clonal β-cells. J. Biol. Chem. 284: 515-524.

CHROMOSOMAL LOCATION

Genetic locus: Slc25a12 (mouse) mapping to 2 C2.

PRODUCT

ARALAR 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ARALAR shRNA Plasmid (m): sc-141183-SH and ARALAR shRNA (m) Lentiviral Particles: sc-141183-V as alternate gene silencing products.

For independent verification of ARALAR (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-141183A, sc-141183B and sc-141183C.

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

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

ARALAR (B-2): sc-271056 is recommended as a control antibody for monitoring of ARALAR 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 ARALAR gene expression knockdown using RT-PCR Primer: ARALAR (m)-PR: sc-141183-PR (20 µl). 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.

PROTOCOLS

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