

Aspartoacylase siRNA (h): sc-93596

BACKGROUND

Aspartoacylase, also known as ASPA, ACY2 or ASP, is a 313 amino acid protein that is expressed in liver, lung and kidney tissue, as well as in skeletal muscle and in cerebral white matter. Existing as a homodimer, Aspartoacylase functions to catalyze the deacetylation of N-acetylaspartic acid (NAA) (a protein whose hydrolysis is crucial to maintenance of intact white matter) to produce acetate and L-aspartate. Defects in the gene encoding Aspartoacylase are the cause of Canavan disease (CAND), which is a rare neurodegenerative condition that is characterized by white matter vacuolization and demyelination, resulting in a spongy deterioration of brain tissue. CAND is generally characterized by atonia of neck muscles, hypotonia, hyperextension of legs and flexion of arms, blindness, severe mental retardation, megaloccephaly and death.

REFERENCES

1. Kaul, R., et al. 1993. Cloning of the human aspartoacylase cDNA and a common missense mutation in Canavan disease. *Nat. Genet.* 5: 118-123.
2. Kaul, R., et al. 1994. Canavan disease: mutations among Jewish and non-Jewish patients. *Am. J. Hum. Genet.* 55: 34-41.
3. Olsen, T.R., et al. 2002. Two novel aspartoacylase gene (ASPA) missense mutations specific to Norwegian and Swedish patients with Canavan disease. *J. Med. Genet.* 39: e55.
4. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 608034. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Le Coq, J., et al. 2006. Characterization of human aspartoacylase: the brain enzyme responsible for Canavan disease. *Biochemistry* 45: 5878-5884.
6. Hershfield, J.R., et al. 2006. Aspartoacylase is a regulated nuclear-cytoplasmic enzyme. *FASEB J.* 20: 2139-2141.
7. Hershfield, J.R., et al. 2007. Mutational analysis of aspartoacylase: implications for Canavan disease. *Brain Res.* 1148: 1-14.

CHROMOSOMAL LOCATION

Genetic locus: ASPA (human) mapping to 17p13.2.

PRODUCT

Aspartoacylase 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 Aspartoacylase shRNA Plasmid (h): sc-93596-SH and Aspartoacylase shRNA (h) Lentiviral Particles: sc-93596-V as alternate gene silencing products.

For independent verification of Aspartoacylase (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-93596A, sc-93596B and sc-93596C.

RESEARCH USE

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

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

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

Aspartoacylase (D-11): sc-377308 is recommended as a control antibody for monitoring of Aspartoacylase 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 Aspartoacylase gene expression knockdown using RT-PCR Primer: Aspartoacylase (h)-PR: sc-93596-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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