

AADAC siRNA (h): sc-77919

BACKGROUND

The assembly of very-low-density lipoproteins (VLDLs) in the secretory apparatus of the hepatocyte relies on the mobilization of triacylglycerol (TAG) from the cytosolic pool by lipolysis and re-esterification. However, some of the re-esterified TAG products are returned to the cytosolic pool in the liver, which protects vulnerable body tissues from excess lipotoxic non-esterified fatty acids in the plasma. Some of the lipases involved in this process include arylacetamide deacetylase (AADAC) and its related proteins AADACL1 and AADACL2. AADAC, a single pass type II membrane protein of the endoplasmic reticulum, is expressed in hepatocytes, intestinal mucosal cells, pancreas and adrenal gland. It plays an important role in the metabolic activation of arylamine substrates to ultimate carcinogens. AADACL1 hydrolyzes the metabolic intermediate 2-acetyl monoalkylglycerol, and its inactivation results in disruption of ether lipid metabolism in cancer cells and impaired cell migration and tumor growth.

REFERENCES

1. Probst, M.R., Jenö, P. and Meyer, U.A. 1991. Purification and characterization of a human liver arylacetamide deacetylase. *Biochem. Biophys. Res. Commun.* 177: 453-459.
2. Probst, M.R., Beer, M., Beer, D., Jenö, P., Meyer, U.A. and Gasser, R. 1994. Human liver arylacetamide deacetylase. Molecular cloning of a novel esterase involved in the metabolic activation of arylamine carcinogens with high sequence similarity to hormone-sensitive lipase. *J. Biol. Chem.* 269: 21650-21656.
3. Yamazaki, K., Kusano, K., Tadano, K. and Tanaka, I. 1997. Radiation hybrid mapping of human arylacetamide deacetylase (AADAC) locus to chromosome 3. *Genomics* 44: 248-250.
4. Trickett, J.I., Patel, D.D., Knight, B.L., Saggerson, E.D., Gibbons, G.F. and Pease, R.J. 2001. Characterization of the rodent genes for arylacetamide deacetylase, a putative microsomal lipase, and evidence for transcriptional regulation. *J. Biol. Chem.* 276: 39522-39532.

CHROMOSOMAL LOCATION

Genetic locus: AADAC (human) mapping to 3q25.1.

PRODUCT

AADAC 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 AADAC shRNA Plasmid (h): sc-77919-SH and AADAC shRNA (h) Lentiviral Particles: sc-77919-V as alternate gene silencing products.

For independent verification of AADAC (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-77919A, sc-77919B and sc-77919C.

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

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

AADAC (G-12): sc-390591 is recommended as a control antibody for monitoring of AADAC 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 AADAC gene expression knockdown using RT-PCR Primer: AADAC (h)-PR: sc-77919-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.