SANTA CRUZ BIOTECHNOLOGY, INC.

AACT siRNA (h): sc-40944

BACKGROUND

The serine proteinase inhibitors (serpins) are a superfamily of proteins with a diverse set of functions, including the control of blood coagulation, complement activation, programmed cell death and development. The most abundant serpins in human plasma are α-1-antitrypsin (AAT) and α-1-antichymotrypsin (AACT). AACT (also called A1AC and SERPINA3) is a plasma protease inhibitor synthesized in the liver as a single glycopeptide chain. In human, the normal serum level of AACT is about one-tenth that of AAT, with which it shares nucleic acid and protein sequence homology. Both are major acute phase reactants; their concentrations in plasma increase in response to trauma, surgery and infection. Elevated levels of AACT are widely, but not universally, reported in the cerebrospinal fluid and plasma of AD patients. Prostate-specific antigen (PSA) and its SDS-stable complex with AACT are in widespread use as markers for the diagnosis of prostate cancer. AACT deficiency may also be a possible cause of chronic liver disease.

REFERENCES

1. Miyake, H., et al. 2001. Value of prostate specific antigen α-1-antichymotrypsin complex for the detection of prostate cancer in patients with a PSA level of 4.1-10.0 ng/mL: comparison with PSA-related parameters. Int. J. Urol. 8: 589-593.
2. Kalsheker, N., et al. 2002. Gene regulation of the serine proteinase inhibitors α-1-antitrypsin and α-1-antichymotrypsin. Biochem. Soc. Trans. 30: 93-98.
3. Yoon, D., et al. 2002. Role of α-1-antichymotrypsin deficiency in promoting cirrhosis in two siblings with heterozygous α-1-antitrypsin deficiency phenotype SZ. Gut 50: 730-732.
4. Wang, X., et al. 2002. Distribution of plasma α-1-antichymotrypsin levels in Alzheimer disease patients and controls and their genetic controls. Neurobiol. Aging 23: 377-382.
5. Hsieh, M.C., et al. 2002. Inhibition of prostate-specific antigen (PSA) by α-1-antichymotrypsin: salt-dependent activation mediated by a conform-ational change. Biochemistry 41: 2990-2997.
6. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 107280. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: SERPINA3 (human) mapping to 14q32.13.

PRODUCT

AACT 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 AACT shRNA Plasmid (h): sc-40944-SH and AACT shRNA (h) Lentiviral Particles: sc-40944-V as alternate gene silencing products.

For independent verification of AACT (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-40944A, sc-40944B and sc-40944C.

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

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

AACT (F-10): sc-518261 is recommended as a control antibody for monitoring of AACT 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) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) 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 AACT gene expression knockdown using RT-PCR Primer: AACT (h)-PR: sc-40944-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.