SANTA CRUZ BIOTECHNOLOGY, INC.

MAAI siRNA (h): sc-40729

BACKGROUND

In humans, maleylacetoacetate isomerase (MAAI, also known as GSTZ1-1) catalyzes the conversion of maleylacetoacetate to fumarylacetoacetate, the fifth step in the phenylalanine/phenylacetate degradation pathway. Deficiencies in other steps of this pathway cause metabolic diseases, including type I tyrosinemia and phenylketonuria. The glutathione S-transferases (GSTs) are found in all aerobic organisms and catalyze the conjugation of glutathione to a wide variety of electrophilic substrates. By sequence alignment and phylogenetic analysis, a new subgroup of GST-like proteins from human, *C. elegans*, and carnation were identified. Human MAAI is 38% and 49% identical to the carnation and *C. elegans* proteins, respectively. Recombinant human MAAI is a dimer. The enzyme exhibits limited activity with known GST substrates. Western blot analysis indicates that MAAI is most abundant in liver, with lower levels detected in skeletal muscle and brain. The gene which encodes MAAI maps to human chromosome 14q24.3.

REFERENCES

1. Berger, R., et al. 1988. Tyrosinemia type Ib caused by maleylacetoacetate isomerase deficiency: a new enzyme defect. Pediat. Res. 23: 328A.
2. Board, P.G., et al. 1997. ζ, a novel class of glutathione transferases in a range of species from plants to humans. Biochem. J. 328: 929-935.
3. Blackburn, A.C., et al. 1998. Characterization and chromosome location of the gene GSTZ1 encoding the human ζ class glutathione transferase and maleylacetoacetate isomerase. Cytogenet. Cell Genet. 83: 109-114.
4. Fernandez-Canon, J.M., et al. 1998. Characterization of a fungal maleylacetoacetate isomerase gene and identification of its human homologue. J. Biol. Chem. 273: 329-337.
5. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 603758. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: GSTZ1 (human) mapping to 14q24.3.

PRODUCT

MAAI 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 MAAI shRNA Plasmid (h): sc-40729-SH and MAAI shRNA (h) Lentiviral Particles: sc-40729-V as alternate gene silencing products.

For independent verification of MAAI (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-40729A, sc-40729B and sc-40729C.

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

MAAI siRNA (h) is recommended for the inhibition of MAAI 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 60 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

MAAI (H-1): sc-271411 is recommended as a control antibody for monitoring of MAAI 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 MAAI gene expression knockdown using RT-PCR Primer: MAAI (h)-PR: sc-40729-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.