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SANTA CRUZ BIOTECHNOLOGY, INC.

MAT Iα siRNA (m): sc-149291

BACKGROUND

Methionine adenosyltransferase (MAT) catalyzes the formation of S-adenosyltransferase (AdoMet) for methionine catabolism in the liver. Two different genes, MAT1A and MAT2A, encode a liver specific and non-liver specific form of MAT, designated MAT Iα and MAT IIα, respectively. Inactivation of the liver specific gene product, designated MAT I/III, associates with liver diseases such as cirrhosis. MAT Iα expression also correlates with a differentiated phenotype, whereas liver cells expressing MAT IIα present a dedifferentiated phenotype and lowered AdoMet synthesis. Likewise, NFκB and TNFα cause a switch from MAT Iα to MAT IIα expression in human hepatocellular carcinoma (HCC), which facilitates cancer cell growth.

REFERENCES

1. Okada, G., et al. 1981. Multiple species of mammalian S-adenosylmethionine synthetase. Partial purification and characterization. Biochemistry 20: 934-940.
2. LeGros, H.L., et al. 2000. Cloning, expression, and functional characterization of the β regulatory subunit of human methionine adenosyltransferase (MAT II). J. Biol. Chem. 275: 2359-2366.
3. LeGros, L., et al. 2001. Regulation of the human MAT2B gene encoding the regulatory β subunit of methionine adenosyltransferase, MAT II. J. Biol. Chem. 276: 24918-24924.
4. Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 605527. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
5. Martínez-Chantar, M.L., et al. 2003. Methionine adenosyltransferase II β subunit gene expression provides a proliferative advantage in human hepatoma. Gastroenterology 124: 940-948.
6. Yang, H., et al. 2008. Expression pattern, regulation, and functions of methionine adenosyltransferase 2β splicing variants in hepatoma cells. Gastroenterology 134: 281-291.
7. Ramani, K., et al. 2008. Leptin's mitogenic effect in human liver cancer cells requires induction of both methionine adenosyltransferase 2A and 2β. Hepatology 47: 521-531.

CHROMOSOMAL LOCATION

Genetic locus: Mat1a (mouse) mapping to 14 B.

PRODUCT

MAT Iα 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 MAT Iα shRNA Plasmid (m): sc-149291-SH and MAT Iα shRNA (m) Lentiviral Particles: sc-149291-V as alternate gene silencing products.

For independent verification of MAT Iα (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-149291A, sc-149291B and sc-149291C.

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

MAT Iα siRNA (m) is recommended for the inhibition of MAT Iα 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

MAT Iα/IIα (B-10): sc-166452 is recommended as a control antibody for monitoring of MAT Iα 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 MAT Iα gene expression knockdown using RT-PCR Primer: MAT Iα (m)-PR: sc-149291-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.