SANTA CRUZ BIOTECHNOLOGY, INC.

β-glucuronidase siRNA (r): sc-270487



BACKGROUND

The enzyme β -glucuronidase catalyzes the conversion of β -D-glucuronoside and water to an alcohol and D-glucuronate. Deficiency of β -glucuronidase is the cause of the human lysosomal storage disorder mucopolysaccharidosis type VII (MPS VII). Specifically, two residues appear important for catalytic activity: Glu 451 and Glu 540. Mutations at these sites affect the overall structure of the protein, which normally consists of a homotetramer with each promoter including a jelly roll barrel, an immunoglobulin constant domain and a TIM barrel. Regulation of β -glucuronidase activity may play a role in tumorigenesis and the invasiveness of a number of cancers, and is also an important factor in the development of functional prodrugs that require the cleavage of an active cytostatic by endogenous enzymes for antitumor activity.

REFERENCES

- 1. Himeno Mnishimura, Y., Tsuji, H. and Kato, K. 1976. Purification and characterization of microsomal and lysosomal β-glucuronidase from rat liver by use of immunoaffinity chromatography. Eur. J. Biochem. 70: 349-359.
- 2. Gupta, G.S. and Singh, G.P. 1983. Isolation and characterization of the major form of β -glucuronidase from human seminal plasma. Biochim. Biophys. Acta 748: 398-404.
- 3. Varma, R., Michos, G.A., Mesmer, R.E., Varma, R.S. and Shirey, R.E. 1983. β-glucuronidase in sera of patients with epileptic seizure activity, diabetes and some other disease states. Neurosci. Lett. 39: 105-111.
- 4. Guise, K.S., Korneluk, R.G., Waye, J., Lamhonwah, A.M., Quan, F., Palmer, R., Ganschow, R.E., Sly, W.S. and Gravel, R.A. 1985. Isolation and expression in *Escherichia coli* of a cDNA clone encoding human β -glucuronidase. Gene 34: 105-110.
- 5. Watson, G., Felder, M., Rabinow, L., Moore, K., Labarca, C., Tietze, C., Vander Molen, G., Bracey, L., Brabant, M., Cai, J. and Paigen, K. 1985. Properties of rat and mouse β -glucuronidase mRNA and cDNA, including evidence for sequence polymorphism and genetic regulation of mRNA levels. Gene 36: 15-25.
- 6. Jain, S., Drendel, W.B., Chen, Z.W., Mathews, F.S., Sly, W.S. and Grubb, J.H. 1996. Structure of human β-glucuronidase reveals candidate lysosomal targeting and active-site motifs. Nat. Struct. Biol. 3: 375-381.
- 7. Vervoort, R., Gitzelmann, R., Bosshard, N., Maire, I., Liebaers, I. and Lissens, W. 1998. Low β-glucuronidase enzyme activity and mutations in the human β-glucuronidase gene in mild mucopolysaccharidosis type VII, pseudodeficiency and a heterozygote. Hum. Genet. 102: 69-78.
- 8. Kurokawa, H., Katsube, K., Podyma, K.A., Ikuta, M., Iseki, H., Nakajima, M., Akashi, T., Omura, K., Takagi, M. and Yanagishita, M. 2003. Heparanase and tumor invasion patterns in human oral squamous cell carcinomaxenografts. Cancer Sci. 94: 277-285.
- 9. Grube, M., Kunert-Keil, C., Sperker, B. and Kroemer, H.K. 2003. Verapamil regulates activity and mRNA-expression of human β -glucuronidase in Hep G2 cells. Naunyn Schmiedebergs Arch. Pharmacol. 368: 463-469.

CHROMOSOMAL LOCATION

Genetic locus: Gusb (rat) mapping to 12q12.

PRODUCT

 β -glucuronidase siRNA (r) 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 β -glucuronidase shRNA Plasmid (r): sc-270487-SH and β-glucuronidase shRNA (r) Lentiviral Particles: sc-270487-V as alternate gene silencing products.

For independent verification of β -glucuronidase (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270487A, sc-270487B and sc-270487C.

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

 β -glucuronidase siRNA (r) is recommended for the inhibition of β-glucuronidase expression in rat 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor β -glucuronidase gene expression knockdown using RT-PCR Primer: β-glucuronidase (r)-PR: sc-270487-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.