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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

β-glucosidase siRNA (m): sc-44905

BACKGROUND

β-glucosidase is a predominantly liver enzyme which efficiently hydrolyzes β-D-glucoside and β-D-galactoside. Defects in β-glucosidase cause Gaucher disease, an inherited condition distinguished by the accumulation of glucosylceramide within the cells of the reticuloendothelial system. β-glucosidase is used in enzyme replacement treatment aimed at treating Gaucher disease. The absorption of dietary flavonoid glycosides in humans involves a critical deglycosylation step that is mediated by epithelial β-glucosidases.

REFERENCES

- Overkleeft, H.S., et al. 1998. Generation of specific deoxynojirimycin-type inhibitors of the non-lysosomal glucosylceramidase. *J. Biol. Chem.* 273: 26522-26527.
- de Graaf, M., et al. 2001. Cloning and characterization of human liver cytosolic β-glycosidase. *Biochem. J.* 356: 907-910.
- Zhao, L., et al. 2003. β-glucosylation as a part of self-resistance mechanism in methymycin/pikromycin producing strain *Streptomyces venezuelae*. *Biochemistry* 42: 14794-14804.
- Nemeth K., et al. 2003. Deglycosylation by small intestinal epithelial cell β-glucosidases is a critical step in the absorption and metabolism of dietary flavonoid glycosides in humans. *Eur. J. Nutr.* 42: 29-42.
- Salvioli, R., et al. 2004. Glucosylceramidase mass and subcellular localization are modulated by cholesterol in Niemann-Pick disease type C. *J. Biol. Chem.* 279: 17674-17680.
- Paal, K., et al. 2004. *Paenibacillus* sp. TS12 glucosylceramidase: kinetic studies of a novel sub-family of family 3 glycosidases and identification of the catalytic residues. *Biochem. J.* 378: 141-149.

CHROMOSOMAL LOCATION

Genetic locus: Gba (mouse) mapping to 3 F1.

PRODUCT

β-glucosidase 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 β-glucosidase shRNA Plasmid (m): sc-44905-SH and β-glucosidase shRNA (m) Lentiviral Particles: sc-44905-V as alternate gene silencing products.

For independent verification of β-glucosidase (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-44905A, sc-44905B and sc-44905C.

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.

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-glucosidase siRNA (m) is recommended for the inhibition of β-glucosidase 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

β-glucosidase (B-6): sc-166407 is recommended as a control antibody for monitoring of β-glucosidase 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 β-glucosidase gene expression knockdown using RT-PCR Primer: β-glucosidase (m)-PR: sc-44905-PR (20 μl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.