SANTA CRUZ BIOTECHNOLOGY, INC.

Glucosidase IIβ siRNA (h): sc-29598

BACKGROUND

Trimming of glucoses from N-linked core glycans on newly synthesized glycoproteins occurs sequentially through the action of Glucosidases I and II in the endoplasmic reticulum (ER). Glucosidase II is an ER-localized enzyme that contains α and β subunits (Glucosidase IIα and Glucosidase IIβ). The α and β subunits form a defined heterodimeric complex. Glucosidase IIα is the catalyitc core of the enzyme and can function independently of the β subunit. The sequence of Glucosidase IIβ encodes protein rich in glutamic and aspartic acid with a putative ER retention signal (HDEL) at the C terminus. The phosphorylated form of Glucosidase IIβ is localized in the plasma membrane and is highly expressed in FGF stimulated fibroblasts and epidermal carcinoma cells. Glucosidase IIβ was first purified from a human carcinoma cell line as a potential substrate for protein kinase C. Through the HDEL signal at the C-terminus, Glucosidase IIβ retains the complete complex in the ER.

REFERENCES

1. Shailubhai, K., et al. 1987. Purification and characterization of Glucosidase I involved in N-linked glycoprotein processing in bovine mammary gland. Biochem. J. 247: 555-562.
2. Saxena, S., et al. 1987. Purification and characterization of Glucosidase II involved in N-linked glycoprotein processing in bovine mammary gland. Biochem. J. 247: 563-570.
3. Trombetta, E.S., et al. 1996. Endoplasmic reticulum Glucosidase II is composed of a catalytic subunit, conserved from yeast to mammals, and a tightly bound noncatalytic HDEL-containing subunit. J. Biol. Chem. 271: 27509-27516.

CHROMOSOMAL LOCATION

Genetic locus: PRKCSH (human) mapping to 19p13.2.

PRODUCT

Glucosidase IIβ 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 Glucosidase IIβ shRNA Plasmid (h): sc-29598-SH and Glucosidase IIβ shRNA (h) Lentiviral Particles: sc-29598-V as alternate gene silencing products.

For independent verification of Glucosidase IIβ (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-29598A, sc-29598B and sc-29598C.

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

Glucosidase IIβ siRNA (h) is recommended for the inhibition of Glucosidase IIβ 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

Glucosidase IIβ (H-4): sc-374457 is recommended as a control antibody for monitoring of Glucosidase IIβ 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 Glucosidase IIβ gene expression knockdown using RT-PCR Primer: Glucosidase IIβ (h)-PR: sc-29598-PR (20 µl, 494 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Yoo, J.J., et al. 2016. Differential sensitivity of hepatocellular carcinoma cells to suppression of hepatocystin transcription under hypoxic conditions. J. Bioenerg. Biomembr. 48: 581-590.
2. Khaodee, W., et al. 2017. Glucosidase IIβ subunit (GluIIβ) plays a role in autophagy and apoptosis regulation in lung carcinoma cells in a p53-dependent manner. Cell. Oncol. 40: 579-591.

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.