

# Glucosidase II $\beta$ siRNA (m): sc-29599

## 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  $\alpha$  and  $\beta$  subunits (Glucosidase II $\alpha$  and Glucosidase II $\beta$ ). The  $\alpha$  and  $\beta$  subunits form a defined heterodimeric complex. Glucosidase II $\alpha$  is the catalytic core of the enzyme and can function independently of the  $\beta$  subunit. The sequence of Glucosidase II $\beta$  encodes protein rich in glutamic and aspartic acid with a putative ER retention signal (HDEL) at the C terminus. The phosphorylated form of Glucosidase II $\beta$  is localized in the plasma membrane and is highly expressed in FGF stimulated fibroblasts and epidermal carcinoma cells. Glucosidase II $\beta$  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 II $\beta$  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.
4. Tremblay, K., et al. 2000. The  $\alpha$ - and  $\beta$ -subunits are required for expression of catalytic activity in the hetero-dimeric Glucosidase II complex from human liver. *Glycobiology* 10: 493-502.
5. Trombetta, E.S., et al. 2001. Quaternary and domain structure of glycoprotein processing Glucosidase II. *Biochemistry* 40: 10717-10822.

## CHROMOSOMAL LOCATION

Genetic locus: Prkcsb (mouse) mapping to 9 A3.

## PRODUCT

Glucosidase II $\beta$  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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Glucosidase II $\beta$  shRNA Plasmid (m): sc-29599-SH and Glucosidase II $\beta$  shRNA (m) Lentiviral Particles: sc-29599-V as alternate gene silencing products.

For independent verification of Glucosidase II $\beta$  (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-29599A, sc-29599B and sc-29599C.

## PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

Glucosidase II $\beta$  siRNA (m) is recommended for the inhibition of Glucosidase II $\beta$  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  $\mu$ M in 66  $\mu$ l. 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 II $\beta$  (H-4): sc-374457 is recommended as a control antibody for monitoring of Glucosidase II $\beta$  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-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 II $\beta$  gene expression knockdown using RT-PCR Primer: Glucosidase II $\beta$  (m)-PR: sc-29599-PR (20  $\mu$ l). 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.