SANTA CRUZ BIOTECHNOLOGY, INC.

H^+/K^+ ATPase β siRNA (m): sc-145846

BACKGROUND

The gastric H^+/K^+ ATPase exists as a heterodimer consisting of an α and a β subunit that work in tandem to transport protons across plasma membranes. H^+/K^+ ATPase β, also known as ATP4B or ATP6B, is a 291 amino acid single-pass type II membrane protein that functions as the β subunit of the H^+/K^+ ATPase heterodimer. Working with the α subunit, H^+/K^+ ATPase β effectively catalyzes the the hydrolysis of ATP coupled with the exchange of H^+ and K^+ ions across the plasma membrane and plays an essential role in gastric acid secretion. The gene encoding H^+/K^+ ATPase β maps to human chromosome 13, which houses over 400 genes, such as BRCA2 and RB1, and comprises nearly 4% of the human genome. Trisomy 13, also known as Patau syndrome, is deadly and the few who survive past one year suffer from permanent neurologic defects, difficulty eating and vulnerability to serious respiratory infections.

REFERENCES

1. Maeda, M., et al. 1990. Human gastric (H^+ + K^+)-ATPase gene. Similarity to (Na^+ + K^+)-ATPase genes in exon/intron organization but difference in control region. J. Biol. Chem. 265: 9027-9032.
2. Ma, J.Y., et al. 1991. cDNA cloning of the β-subunit of the human gastric H,K-ATPase. Biochem. Biophys. Res. Commun. 180: 39-45.
3. Song, I., et al. 1992. Mapping of the gene encoding the β-subunit of H^+,K^+-ATPase to human chromosome 13q34 by fluorescence *in situ* hybridization. Genomics 14: 1114-1115.
4. Callaghan, J.M., et al. 1995. Renal expression of the gene encoding the gastric H^+-K^+-ATPase β-subunit. Am. J. Physiol. 268: F363-F374.
5. Sachs, G. 1997. Proton pump inhibitors and acid-related diseases. Pharmacotherapy 17: 22-37.
6. Asano, S., et al. 2000. The roles of carbohydrate chains of the β-subunit on the functional expression of gastric H^+,K^+-ATPase. J. Biol. Chem. 275: 8324-8330.
7. Asano, S., et al. 2004. Molecular and cellular regulation of the gastric proton pump. Biol. Pharm. Bull. 27: 1-12.

CHROMOSOMAL LOCATION

Genetic locus: Atp4b (mouse) mapping to 8 A1.1.

PRODUCT

H^+/K^+ ATPase β 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 H^+/K^+ ATPase β shRNA Plasmid (m): sc-145846-SH and H^+/K^+ ATPase β shRNA (m) Lentiviral Particles: sc-145846-V as alternate gene silencing products.

For independent verification of H^+/K^+ ATPase β (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-145846A, sc-145846B and sc-145846C.

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

H^+/K^+ ATPase β siRNA (m) is recommended for the inhibition of H^+/K^+ ATPase β 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

H^+/K^+ ATPase β (C-4): sc-374094 is recommended as a control antibody for monitoring of H^+/K^+ ATPase β 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 H^+/K^+ ATPase β gene expression knockdown using RT-PCR Primer: H^+/K^+ ATPase β (m)-PR: sc-145846-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.