SANTA CRUZ BIOTECHNOLOGY, INC.

ATP6F siRNA (h): sc-78935

BACKGROUND

Vacuolar-type H^+-ATPase (V-ATPase) is a multisubunit enzyme responsible for acidification of eukaryotic intracellular organelles. V-ATPase-dependent organelle acidification is essential for intracellular processes such as protein sorting, zymogen activation, and receptor-mediated endocytosis. ATP6F, also known as ATP6V0B or V-type proton ATPase 21 kDa proteolipid subunit, is a 205 amino acid mulit-pass membrane protein that belongs to the V-ATPase proteolipid subunit family. ATP6F contains five transmembrane segments and a conserved glutamic acid residue that participates in proton transport activity. ATP6F is ubiquitously expressed and localizes to vacuole. The ATP6F gene contains eight exons and spans approximately 4 kb. The ATP6V0B gene is conserved in canine, bovine, mouse, rat, zebrafish, fruit fly, mosquito, *C. elegans*, *S. pombe*, *S. cerevisiae*, *K. lactis*, *E. gossypii*, *M. grisea*, *N. crassa*, *A. thaliana*, rice and *P. falciparum*, and maps to human chromosome 1p34.1.

REFERENCES

1. Stevens, T.H. and Forgac, M. 1997. Structure, function and regulation of the vacuolar (H^+)-ATPase. Annu. Rev. Cell Dev. Biol. 13: 779-808.
2. Finbow, M.E. and Harrison, M.A. 1997. The vacuolar H^+-ATPase: a universal proton pump of eukaryotes. Biochem. J. 324: 697-712.
3. Oka, T., et al. 1997. Three vha genes encode proteolipids of Caenorhabditis elegans vacuolar-type ATPase. Gene structures and preferential expression in an H-shaped excretory cell and rectal cells. J. Biol. Chem. 272: 24387-24392.
4. Nishigori, H., et al. 1998. Identification and characterization of the gene encoding a second proteolipid subunit of human vacuolar H^+-ATPase (ATP6F). Genomics 50: 222-228.
5. Forgac, M. 1999. Structure and properties of the vacuolar (H^+)-ATPases. J. Biol. Chem. 274: 12951-12954.
6. Online Mendelian Inheritance in Man, OMIM™. 1999. Johns Hopkins University, Baltimore, MD. MIM Number: 603717. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
7. Nishi, T. and Forgac, M. 2002. The vacuolar (H^+)-ATPases—nature's most versatile proton pumps. Nat. Rev. Mol. Cell Biol. 3: 94-103.
8. Morel, N. 2003. Neurotransmitter release: the dark side of the vacuolar-H^+ATPase. Biol. Cell 95: 453-457.
9. Rojas, J.D., et al. 2004. Plasmalemmal V-H^+-ATPases regulate intracellular pH in human lung microvascular endothelial cells. Biochem. Biophys. Res. Commun. 320: 1123-1132.

CHROMOSOMAL LOCATION

Genetic locus: ATP6V0B (human) mapping to 1p34.1.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

PRODUCT

ATP6F 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 ATP6F shRNA Plasmid (h): sc-78935-SH and ATP6F shRNA (h) Lentiviral Particles: sc-78935-V as alternate gene silencing products.

For independent verification of ATP6F (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-78935A, sc-78935B and sc-78935C.

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

ATP6F siRNA (h) is recommended for the inhibition of ATP6F 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ATP6F gene expression knockdown using RT-PCR Primer: ATP6F (h)-PR: sc-78935-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.