SANTA CRUZ BIOTECHNOLOGY, INC.

Na^+/K^+-ATPase β1 siRNA (h2): sc-270380

BACKGROUND

The ubiquitously expressed sodium/potassium-ATPase (Na^+/K^+-ATPase) exists as a oligomeric plasma membrane complex that couples the hydrolysis of one molecule of ATP to the importation of three Na^+ ions and two K^+ ions against their respective electrochemical gradients. As a member of the P-type family of ion motives, Na^+/K^+-ATPase plays a critical role in maintaining cellular volume, resting membrane potential and Na^+-coupled solute transport. Multiple isoforms of three subunits, α, β and γ, comprise the Na^+/K^+-ATPase oligomer. The α subunit contains the binding sites for ATP and the cations; the glycosylated β subunit ensures correct folding and membrane insertion of the α subunits. The small γ subunit co-localizes with the α subunit in nephron segments, where it increases the affinity of Na^+/K^+-ATPase for ATP. The β subunit, but not the γ subunit, is essential for normal activity of Na^+/K^+-ATPase.

REFERENCES

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2. Ackermann, U., et al. 1990. Mutual dependence of Na,K-ATPase α- and β-subunits for correct post-translational processing and intracellular transport. FEBS Lett. 269: 105-108.
3. McDonough, A.A., et al. 1990. The sodium pump needs its β subunit. FASEB J. 4: 1598-1605.
4. Pedemonte, C.H., et al. 1990. Chemical modification as an approach to elucidation of sodium pump structure-function relations. Am. J. Physiol. 258: C1-C23.
5. Mercer, R.W., et al. 1993. Molecular cloning and immunological chracterization of the γ-polypeptide, a small protein associated with Na,K-ATPase. J. Cell Biol. 121: 579-586.
6. DeTomaso, A.W., et al. 1993. Expression, targeting, and assembly of functional Na,K-ATPase polypeptides in baculovirus-infected insect cells. J. Biol. Chem. 268: 1470-1478.
7. Scheiner-Bobis, G., et al. 1994. Subunit requirements for expression of functional sodium pumps in yeast cells. Biochim. Biophys. Acta 1193: 226-234.

CHROMOSOMAL LOCATION

Genetic locus: ATP1B1 (human) mapping to 1q24.2.

PRODUCT

Na^+/K^+-ATPase β1 siRNA (h2) 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 Na^+/K^+-ATPase β1 shRNA Plasmid (h2): sc-270380-SH and Na^+/K^+-ATPase β1 shRNA (h2) Lentiviral Particles: sc-270380-V as alternate gene silencing products.

For independent verification of Na^+/K^+-ATPase β1 (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270380A, sc-270380B and sc-270380C.

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

Na^+/K^+-ATPase β1 siRNA (h2) is recommended for the inhibition of Na^+/K^+-ATPase β1 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

Na^+/K^+-ATPase β1 (E-4): sc-376406 is recommended as a control antibody for monitoring of Na^+/K^+-ATPase β1 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 Na^+/K^+-ATPase β1 gene expression knockdown using RT-PCR Primer: Na^+/K^+-ATPase β1 (h2)-PR: sc-270380-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.