Na $^+$ /K $^+$ -ATPase β 1 siRNA (m): sc-36009



The Power to Question

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.
- 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.
- 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.

CHROMOSOMAL LOCATION

Genetic locus: Atp1b1 (mouse) mapping to 1 H2.2.

PRODUCT

Na+/K+-ATPase β 1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Na+/K+-ATPase β 1 shRNA Plasmid (m): sc-36009-SH and Na+/K+-ATPase β 1 shRNA (m) Lentiviral Particles: sc-36009-V as alternate gene silencing products.

For independent verification of Na+/K+-ATPase $\beta1$ (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-36009A, sc-36009B and sc-36009C.

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 μ l of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNAse-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Na+/K+-ATPase β 1 siRNA (m) is recommended for the inhibition of Na+/K+-ATPase β 1 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

Na+/K+-ATPase β 1 (C464.8): sc-21713 is recommended as a control antibody for monitoring of Na+/K+-ATPase β 1 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz $^{\circ}$ Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz $^{\circ}$ Mounting Medium: sc-24941 or UltraCruz $^{\circ}$ Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Na+/K+-ATPase β 1 gene expression knockdown using RT-PCR Primer: Na+/K+-ATPase β 1 (m)-PR: sc-36009-PR (20 μ I, 595 bp). 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.