SANTA CRUZ BIOTECHNOLOGY, INC.

FXYD2 siRNA (h): sc-42422

BACKGROUND

The human FXYD2 (pronounced fix-id) gene maps to chromosome 11q23.3 and encodes the γ subunit of Na,K-ATPase (NKA). FXYD2 γ subunit modulates NKA activity by inducing ion channel activity. The mammalian FXYD family maintains Na^+ and K^+ gradients between the intracellular and extracellular milieus of cells in processes such as renal Na^+-reabsorption, muscle contraction, and neuronal excitability. FXYDs are single-span membrane proteins that share a 35 amino acid signature domain, beginning with the sequence PFXYD and containing 7 invariant and 6 conserved amino acids. Other members of the FXYD family include FXYD1 (PLM, phospholemman), FXYD3 (Mat8, mammary tumor protein 8), FXYD4 (CHIF), and FXYD5 (RIC).

REFERENCES

1. Sweadner, K.J., et al. 2000. The FXYD gene family of small ion transport regulators or channels: cDNA sequence, protein signature sequence, and expression. Genomics 68: 41-56.
2. Beguin, P., et al. 2001. CHIF, a member of the FXYD protein family, is a regulator of Na,K-ATPase distinct from the γ-subunit. EMBO J. 20: 3993-4002.
3. Crambert, G., et al. 2002. Phospholemman (FXYD1) associates with Na,K-ATPase and regulates its transport properties. Proc. Natl. Acad. Sci. USA 99: 11476-11481.
4. Crambert, G., et al. 2003. FXYD proteins: new tissue-specific regulators of the ubiquitous Na,K-ATPase. Sci. STKE 2003: RE1.
5. Crowell, K.J., et al. 2003. Expression and characterization of the FXYD ion transport regulators for NMR structural studies in lipid micelles and lipid bilayers. Biochim. Biophys. Acta 1645: 15-21.
6. Lindzen, M., et al. 2003. Structure-function relations of interactions between Na/K-ATPase, the γ subunit and corticosteroid hormone-induced factor. J. Biol. Chem. 278: 18738-18743.
7. LocusLink Report (LocusID: 486). http://www.ncbi.nlm.nih.gov/LocusLink/

CHROMOSOMAL LOCATION

Genetic locus: FXYD2 (human) mapping to 11q23.3.

PRODUCT

FXYD2 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 FXYD2 shRNA Plasmid (h): sc-42422-SH and FXYD2 shRNA (h) Lentiviral Particles: sc-42422-V as alternate gene silencing products.

For independent verification of FXYD2 (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-42422A, sc-42422B and sc-42422C.

PROTOCOLS

See our web site at 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 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

FXYD2 siRNA (h) is recommended for the inhibition of FXYD2 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

FXYD2 (ZW-5): sc-81876 is recommended as a control antibody for monitoring of FXYD2 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 FXYD2 gene expression knockdown using RT-PCR Primer: FXYD2 (h)-PR: sc-42422-PR (20 µl, 447 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.