

ANO4 siRNA (m): sc-154402

BACKGROUND

ANO4 (anoctamin 4), also known as TMEM16D (transmembrane protein 16D), is a 955 amino acid multi-pass membrane protein. ANO4 is encoded by a gene that maps to chromosome 12 and is expressed as three isoforms due to alternative splicing events. Encoding over 1,100 genes within 132 million bases, chromosome 12 makes up about 4.5% of the human genome. A number of skeletal deformities are linked to chromosome 12, including hypochondrogenesis, achondrogenesis and Kniest dysplasia. Noonan syndrome, which is characterized by heart and facial developmental defects, is caused by a mutant form of the PTPN11 gene product, SH-PTP2. Chromosome 12 is also home to a homeobox gene cluster which encodes crucial transcription factors for morphogenesis, and the natural killer complex gene cluster encoding C-type lectin proteins which mediate the NK cell response to MHC I interaction.

REFERENCES

1. Katoh, M. and Katoh, M. 2003. FLJ10261 gene, located within the CCND1-EMS1 locus on human chromosome 11q13, encodes the eight-transmembrane protein homologous to C12orf3, C11orf25 and FLJ34272 gene products. *Int. J. Oncol.* 22: 1375-1381.
2. Katoh, M. and Katoh, M. 2004. Identification and characterization of TMEM16E and TMEM16F genes in silico. *Int. J. Oncol.* 24: 1345-1349.
3. Zunkeller, W., Volleth, M., Muschke, P., Tönnies, H., Heller, A., Liehr, T., Wieacker, P. and Stumm, M. 2004. Genotype/phenotype analysis in a patient with pure and complete trisomy 12p. *Am. J. Med. Genet. A* 129A: 261-264.
4. Kelley, J., Walter, L. and Trowsdale, J. 2005. Comparative genomics of natural killer cell receptor gene clusters. *PLoS Genet.* 1: e27.
5. Stein, R. 2007. Genetics of Noonan syndrome-a new gene, and the search is still on. *Clin. Genet.* 72: 402-404.
6. Duran, C. and Hartzell, H.C. 2011. Physiological roles and diseases of Tmem16/Anoctamin proteins: are they all chloride channels? *Acta Pharmacol. Sin.* 32: 685-692.
7. Kunzelmann, K., Tian, Y., Martins, J.R., Faria, D., Kongsuphol, P., Ousingsawat, J., Thevenod, F., Roussa, E., Rock, J. and Schreiber, R. 2011. Anoctamins. *Pflugers Arch.* 462: 195-208.
8. Winpenny, J.P. and Gray, M.A. 2012. The anoctamin (TMEM16) gene family: calcium-activated chloride channels come of age. *Exp. Physiol.* 97: 175-176.
9. Tian, Y., Schreiber, R. and Kunzelmann, K. 2012. Anoctamins are a family of Ca²⁺-activated Cl⁻ channels. *J. Cell Sci.* 125: 4991-4998.

CHROMOSOMAL LOCATION

Genetic locus: Ano4 (mouse) mapping to 10 C1.

PROTOCOLS

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

PRODUCT

ANO4 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 ANO4 shRNA Plasmid (m): sc-154402-SH and ANO4 shRNA (m) Lentiviral Particles: sc-154402-V as alternate gene silencing products.

For independent verification of ANO4 (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-154402A, sc-154402B and sc-154402C.

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

ANO4 siRNA (m) is recommended for the inhibition of ANO4 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 μ M in 66 μ l. 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 ANO4 gene expression knockdown using RT-PCR Primer: ANO4 (m)-PR: sc-154402-PR (20 μ l). 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.