

TMEM12 siRNA (h): sc-90745

BACKGROUND

USHSA (Usher syndrome type-3) is thought to be involved in the maintenance of the inner ear and retina, specifically playing a role in excitatory ribbon synapse junctions between hair cells and cochlear ganglion cells, as well as in analogous synapses within the retina. Defects in the gene encoding USH3A are the cause of Usher syndrome type 3 (USH3), a genetically heterogeneous condition characterized by postlingual progressive deafness and onset of retinitis pigmentosa in the second decade of life. TMEM12 (Transmembrane protein 12), also known as USH3A1 or CLRN3, is a 226 amino acid multi-pass membrane protein that belongs to the clarin family. Existing as two alternatively spliced isoforms TMEM12 may display similar functions as USHSA.

REFERENCES

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2. Joensuu, T., et al. 1996. Refined mapping of the Usher syndrome type III locus on chromosome 3, exclusion of candidate genes, and identification of the putative mouse homologous region. *Genomics* 38: 255-263.
3. Adato, A., et al. 1999. Possible interaction between USH1B and USH3 gene products as implied by apparent digenic deafness inheritance. *Am. J. Hum. Genet.* 65: 261-265.
4. Joensuu, T., et al. 2000. A sequence-ready map of the Usher syndrome type III critical region on chromosome 3q. *Genomics* 63: 409-416.
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CHROMOSOMAL LOCATION

Genetic locus: CLRN3 (human) mapping to 10q26.2.

PRODUCT

TMEM12 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TMEM12 shRNA Plasmid (h): sc-90745-SH and TMEM12 shRNA (h) Lentiviral Particles: sc-90745-V as alternate gene silencing products.

For independent verification of TMEM12 (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-90745A, sc-90745B and sc-90745C.

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

TMEM12 siRNA (h) is recommended for the inhibition of TMEM12 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 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.

GENE EXPRESSION MONITORING

TMEM12 (D-2): sc-398948 is recommended as a control antibody for monitoring of TMEM12 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 TMEM12 gene expression knockdown using RT-PCR Primer: TMEM12 (h)-PR: sc-90745-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.

PROTOCOLS

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