SANTA CRUZ BIOTECHNOLOGY, INC.

cadherin-23 siRNA (m): sc-43010

BACKGROUND

Cadherin-23 represents the first in this family of calcium binding proteins of which mutations in the extracellular calcium binding domain contribute to an inherited disorder, Usher syndrome type 1D (USH1D). Patients with USH1D exhibit congenital sensorineural hearing loss, vestibular dysfunction and visual impairment due to early onset of retinitis pigmentosa (RP). In the inner ear, cadherin-23 interacts with Myosin VIIIA and harmonin to form a functional network duing hair cell differentiation and in the retina to assemble a supramolecular complex contributing to the organization of the cytoskeletal matrices of the pre- and post-synaptic region. A number of cadherin-23 splice variants exist in association with various phenotypic expression, indicating that differential mutations result in variable presentation of the disease.

REFERENCES

1. Di Palma, F., et al. 2001. Genomic structure, alternative splice forms and normal and mutant alleles of cadherin-23 (Cdh23). Gene 281: 31-41.
2. Bolz, H., et al. 2001. Mutation of CDH23, encoding a new member of the cadherin gene family, causes Usher syndrome type 1D. Nat. Genet. 27: 108-112.
3. Boëda, B., et al. 2002. Myosin VIIa, harmonin and cadherin-23, three Usher I gene products that cooperate to shape the sensory hair cell bundle. EMBO J. 21: 6689-6699.
4. Siemens, J., et al. 2002. The Usher syndrome proteins cadherin-23 and harmonin form a complex by means of PDZ-domain interactions. Proc. Natl. Acad. Sci. USA 99: 14946-14951.
5. Reiners, J., et al. 2003. Differential distribution of harmonin isoforms and their possible role in Usher-1 protein complexes in mammalian photoreceptor cells. Invest. Ophthalmol. Vis. Sci. 44: 5006-5015.
6. Noben-Trauth, K., et al. 2003. Association of cadherin-23 with polygenic inheritance and genetic modification of sensorineural hearing loss. Nat. Genet. 35: 21-23.
7. de Brouwer, A.P., et al. 2003. Mutations in the calcium-binding motifs of CDH23 and the 35delG mutation inGJB2 cause hearing loss in one family. Hum. Genet. 112: 156-163.

CHROMOSOMAL LOCATION

Genetic locus: Cdh23 (mouse) mapping to 10 B4.

PRODUCT

cadherin-23 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see cadherin-23 shRNA Plasmid (m): sc-43010-SH and cadherin-23 shRNA (m) Lentiviral Particles: sc-43010-V as alternate gene silencing products.

For independent verification of cadherin-23 (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-43010A, sc-43010B and sc-43010C.

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

cadherin-23 siRNA (m) is recommended for the inhibition of cadherin-23 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

cadherin-23 (H-7): sc-166066 is recommended as a control antibody for monitoring of cadherin-23 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 cadherin-23 gene expression knockdown using RT-PCR Primer: cadherin-23 (m)-PR: sc-43010-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.