SANTA CRUZ BIOTECHNOLOGY, INC.

Keratin 25 siRNA (h): sc-93610

BACKGROUND

The keratin multigene family is made of the "soft" epithelial cytokeratins and the "hard" hair keratins. While the epithelial cytokeratins are involved in the layering and formation of epithelia, the hair keratins are responsible for creating nails and hair. There are two types of hair keratins: the acidic type I hair keratin proteins and the basic/neutral type II hair keratin proteins. Keratin 25, also known as KRT25A or KRT25, is a 450 amino acid cytoplasmic protein belonging to the intermediate filament family. Essential for the proper assembly of type I and type II keratin protein complexes, Keratin 25 is strongly expressed in skin and scalp. Keratin 25 exists as a heterotetramer of two type I and two type II keratins and interacts with Cytokeratin 6. Keratin 25 is required for proper formation of keratin intermediate filaments in the inner root sheath (irs).

REFERENCES

1. Rogers, M.A., et al. 1995. Sequence data and chromosomal localization of human type I and type II hair keratin genes. Exp. Cell Res. 220: 357-362.
2. Winter, H., et al. 1997. A new mutation in the type II hair cortex keratin hHb1 involved in the inherited hair disorder monilethrix. Hum. Genet. 101: 165-169.
3. Bowden, P.E., et al. 1998. Characterization and chromosomal localization of human hair-specific keratin genes and comparative expression during the hair growth cycle. J. Invest. Dermatol. 110: 158-164.
4. Winter, H., et al. 1998. A variable monilethrix phenotype associated with a novel mutation, Glu402Lys, in the helix termination motif of the type II hair keratin hHb1. J. Invest. Dermatol. 111: 169-172.
5. Rogers, M.A., et al. 2000. Characterization of a 300 kbp region of human DNA containing the type II hair keratin gene domain. J. Invest. Dermatol. 114: 464-472
6. Coulombe, P.A. and Omary, M.B. 2002. "Hard" and "soft" principles defining the structure, function and regulation of keratin intermediate filaments. Curr. Opin. Cell Biol. 14: 110-122.
7. Langbein, L. and Schweizer, J. 2005. Keratins of the human hair follicle. Int. Rev. Cytol. 243: 1-78.

CHROMOSOMAL LOCATION

Genetic locus: KRT25 (human) mapping to 17q21.2.

PRODUCT

Keratin 25 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 Keratin 25 shRNA Plasmid (h): sc-93610-SH and Keratin 25 shRNA (h) Lentiviral Particles: sc-93610-V as alternate gene silencing products.

For independent verification of Keratin 25 (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-93610A, sc-93610B and sc-93610C.

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

Keratin 25 siRNA (h) is recommended for the inhibition of Keratin 25 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

Keratin 25 (B-2): sc-398320 is recommended as a control antibody for monitoring of Keratin 25 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 Keratin 25 gene expression knockdown using RT-PCR Primer: Keratin 25 (h)-PR: sc-93610-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.