SANTA CRUZ BIOTECHNOLOGY, INC.

TLE5 siRNA (m): sc-63134

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The Power to Question

BACKGROUND

The Notch signaling pathway controls cellular interactions important for the specification of a variety of fates in both vertebrates and invertebrates. Key players in the Notch pathway are the TLE genes (for transducin-like enhancer of split, also designated ESG for enhancer of split groucho), which are human homologs of the *Drosophila* groucho gene. TLE5, also known as AES (amino-terminal enhancer of split), GRG or ESP1, is a 197 amino acid nuclear protein that belongs to the TLE family. Expressed predominately in fetal brain, liver, lung, heart and kidney and in adult muscle, TLE5 functions as either a homo-oligomer or a heterooligomer with other TLE family members and, through this association, dominantly represses the expression of TLE genes. In addition, TLE5 can repress NFκB-regulated gene expression and is thought to play an important role in initiating and maintaining cell differentiation events. Two isoforms of TLE5 exist due to alternative splicing events.

REFERENCES

1. Miyasaka, H., et al. 1993. Molecular cloning and expression of mouse and human cDNA encoding AES and ESG proteins with strong similarity to *Drosophila* enhancer of split groucho protein. Eur. J. Biochem. 216: 343-352.
2. Hou, E.W. and Li, S.S. 1998. Genomic organization and chromosome localization to band 19p13.3 of the human AES gene: gene product exhibits strong similarity to the N-terminal domain of *Drosophila* enhancer of split groucho protein. DNA Cell Biol. 17: 911-913.
3. Tetsuka, T., et al. 2000. Inhibition of nuclear factor-κB-mediated transcription by association with the amino-terminal enhancer of split, a groucho-related protein lacking WD40 repeats. J. Biol. Chem. 275: 4383-4390.
4. Wang, J.C., et al. 2000. Transducin-like enhancer of split proteins, the human homologs of *Drosophila* groucho, interact with hepatic nuclear factor 3β. J. Biol. Chem. 275: 18418-18423.
5. Yochum, G.S. and Ayer, D.E. 2001. Pf1, a novel PHD zinc finger protein that links the TLE corepressor to the mSin3A-histone deacetylase complex. Mol. Cell. Biol. 21: 4110-4118.
6. López-Ríos, J., et al. 2003. Six3 and Six6 activity is modulated by members of the groucho family. Development 130: 185-195.

CHROMOSOMAL LOCATION

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

PRODUCT

TLE5 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 TLE5 shRNA Plasmid (m): sc-63134-SH and TLE5 shRNA (m) Lentiviral Particles: sc-63134-V as alternate gene silencing products.

For independent verification of TLE5 (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-63134A, sc-63134B and sc-63134C.

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

TLE5 siRNA (m) is recommended for the inhibition of TLE5 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

TLE5 (C-7): sc-515756 is recommended as a control antibody for monitoring of TLE5 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 TLE5 gene expression knockdown using RT-PCR Primer: TLE5 (m)-PR: sc-63134-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.