

TFEC siRNA (h): sc-38511

BACKGROUND

The DNA-binding factor TFE3 contains adjacent helix-loop-helix (HLH) and leucine zipper (LZ) domains flanked by an upstream basic region. These protein motifs are frequently observed in other transcription factors and are particularly common to members of the Myc family. TFE3 is ubiquitously expressed and can directly associate with DNA as either homodimers or heterodimers formed with two related proteins, TFEB or TFEC. TFE3 binds to and activates the microE3 motif of the immunoglobulin heavy-chain enhancer to induce B cell-specific gene transcription and DNA recombination. TFEB binds to the major late promoter of adenovirus and specifically associates with DNA as both a homodimer and a heterodimer with TFE3. TFEB is expressed at low levels in the embryo but at high levels in the trophoblast cells of the placenta, where it plays a critical role in regulating normal vascularization of the placenta. TFEC shares a bHLH/LZ structure with TFE3 and a closely related protein microphthalmia-associated transcription factor (MITF), which is critically involved in melanocyte differentiation. Unlike TFE3, the expression of TFEC is largely restricted to fibroblasts, myoblasts, chondrosarcoma cells and myeloma cells.

REFERENCES

1. Beckmann, H., et al. 1990. TFE3: a helix-loop-helix protein that activates transcription through the immunoglobulin enhancer μ E3 motif. *Genes Dev.* 4: 167-179.
2. Fisher, D.E., et al. 1991. TFEB has DNA-binding and oligomerization properties of a unique helix-loop-helix/leucine-zipper family. *Genes Dev.* 5: 2342-2352.
3. Kerkhoff, E., et al. 1991. Sequence-specific DNA binding by Myc proteins. *Proc. Natl. Acad. Sci. USA* 88: 4323-4327.
4. Artandi, S.E., et al. 1994. The basic helix-loop-helix-zipper domain of TFE3 mediates enhancer-promoter interaction. *Mol. Cell. Biol.* 14: 7704-7716.
5. Yasumoto, K., et al. 1997. Molecular cloning of cDNA encoding a human TFEC isoform, a newly identified transcriptional regulator. *Biochim. Biophys. Acta* 1353: 23-31.
6. Steingrimsson, E., et al. 1998. The bHLH-Zip transcription factor TFEB is essential for placental vascularization. *Development* 125: 4607-4616.

CHROMOSOMAL LOCATION

Genetic locus: TFEC (human) mapping to 7q31.2.

PRODUCT

TFEC 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 TFEC shRNA Plasmid (h): sc-38511-SH and TFEC shRNA (h) Lentiviral Particles: sc-38511-V as alternate gene silencing products.

For independent verification of TFEC (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-38511A, sc-38511B and sc-38511C.

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

TFEC siRNA (h) is recommended for the inhibition of TFEC 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

TFEC (E-12): sc-515031 is recommended as a control antibody for monitoring of TFEC 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 TFEC gene expression knockdown using RT-PCR Primer: TFEC (h)-PR: sc-38511-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.