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EBF3 siRNA (h): sc-43743

BACKGROUND

B lymphocyte maturation is an intricate process that requires a distinct set of transcription factors with respect to the stage of cell differentiation and cell lineage. Among the transcriptional regulators involved in the early stages of B cell development, early B cell factor 3 (EBF3) targets promoter elements for B lymphoid kinase (Blk) and genes encoding portions of the early stage B cell receptors (BCR), which are necessary for initiation of Ig light chain gene recombination and Src kinase (Blk) signaling. EBF3 is a nuclear transcription factor that is highly expressed in brain and exists as either a homodimer or a heterodimer with related family members. Specifically, EBF3 exhibits transcriptional activation activity on target genes by interacting with the core DNA sequence, CCCNNGGG, through a DNA-recognition domain containing a zinc-coordination motif. Two isoforms of EBF3 exist due to alternative splicing events.

REFERENCES

1. Wang, M.M., et al. 1993. Molecular cloning of the olfactory neuronal transcription factor OLF1 by genetic selection in yeast. *Nature* 364: 121-126.
2. Lin, H., et al. 1995. Failure of B cell differentiation in mice lacking the transcription factor EBF. *Nature* 376: 263-267.
3. Hagman, J., et al. 1995. EBF contains a novel zinc-coordination motif and multiple dimerization and transcriptional activation domains. *EMBO J.* 14: 2907-2916.
4. Sigvardsson, M., et al. 1997. EBF and E47 collaborate to induce expression of the endogenous immunoglobulin surrogate light chain genes. *Immunity* 7: 25-36.
5. Akerblad, P., et al. 1999. The B29 (immunoglobulin β -chain) gene is a genetic target for early B cell factor. *Mol. Cell. Biol.* 19: 392-401.
6. Akerblad, P., et al. 1999. Early B cell factor is an activator of the B lymphoid kinase promoter in early B cell development. *J. Immunol* 163: 5453-5461.
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CHROMOSOMAL LOCATION

Genetic locus: EBF3 (human) mapping to 10q26.3.

PRODUCT

EBF3 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see EBF3 shRNA Plasmid (h): sc-43743-SH and EBF3 shRNA (h) Lentiviral Particles: sc-43743-V as alternate gene silencing products.

For independent verification of EBF3 (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-43743A, sc-43743B and sc-43743C.

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

EBF3 siRNA (h) is recommended for the inhibition of EBF3 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

EBF3 (N7Q): sc-81999 is recommended as a control antibody for monitoring of EBF3 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 EBF3 gene expression knockdown using RT-PCR Primer: EBF3 (h)-PR: sc-43743-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.