

ATF-6 β siRNA (h): sc-105103

BACKGROUND

ATF-6 is a member of the basic-leucine zipper family of transcription factors. Endoplasmic reticulum stress causes cleavage of transmembrane ATF-6 and translocation of active ATF-6 to the nucleus. Soluble ATF-6 can exist as either an ATF-6 β homodimer or an ATF-6 α / β heterodimer. Binding of the ATF-6 β homodimer or ATF-6 α / β heterodimer to the nuclear transcription factor Y C (NF-YC) induces ER chaperone transcription.

REFERENCES

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3. Wang, Y., Shen, J., Arenzana, N., Tirasophon, W., Kaufman, R.J. and Prywes, R. 2000. Activation of ATF-6 and an ATF-6 DNA binding site by the endoplasmic reticulum stress response. *J. Biol. Chem.* 275: 27013-27020.
4. Yoshida, H., Okada, T., Haze, K., Yanagi, H., Yura, T., Negishi, M. and Mori, K. 2000. ATF-6 activated by proteolysis binds in the presence of NF-Y (CBF) directly to the *cis*-acting element responsible for the mammalian unfolded protein response. *Mol. Cell. Biol.* 20: 6755-6767.
5. Urano, F., Bertolotti, A. and Ron, D. 2000. IRE1 and efferent signaling from the endoplasmic reticulum. *J. Cell Sci.* 113: 3697-3702.
6. Li, M., Baumeister, P., Roy, B., Phan, T., Foti, D., Luo, S. and Lee, A.S. 2000. ATF-6 as a transcription activator of the endoplasmic reticulum stress element: Thapsigargin stress-induced changes and synergistic interactions with NF-Y and YY1. *Mol. Cell. Biol.* 20: 5096-5106.

CHROMOSOMAL LOCATION

Genetic locus: ATF6B (human) mapping to 6p21.33.

PRODUCT

ATF-6 β 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 ATF-6 β shRNA Plasmid (h): sc-105103-SH and ATF-6 β shRNA (h) Lentiviral Particles: sc-105103-V as alternate gene silencing products.

For independent verification of ATF-6 β (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-105103A, sc-105103B and sc-105103C.

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.

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

ATF-6 β siRNA (h) is recommended for the inhibition of ATF-6 β 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

ATF-6 β (4D10): sc-293306 is recommended as a control antibody for monitoring of ATF-6 β 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 ATF-6 β gene expression knockdown using RT-PCR Primer: ATF-6 β (h)-PR: sc-105103-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.