

DnaJB14 siRNA (h): sc-88854

BACKGROUND

The DnaJ family, one of the largest of all the chaperone families, has evolved with diverse cellular localization and functions. The presence of a J domain defines a protein as a member of the DnaJ family. DnaJ heat-shock induced proteins are derived from the bacterium *Escherichia coli* and are under the control of the htpR regulatory protein. DnaJ proteins play a critical role in the HSP 70 chaperone machine by interacting with HSP 70 to stimulate ATP hydrolysis. Members of this family contain cysteine-rich regions that are composed of zinc fingers that form a peptide-binding domain responsible for chaperone function. DnaJ family members are important mediators of proteolysis and are involved in the regulation of protein degradation, exocytosis and endocytosis. DnaJB14 (DnaJ (HSP 40) homolog, subfamily B, member 14) is a 379 amino acid single-pass membrane protein containing one J domain and is thought to act as a co-chaperone. DnaJB14 exists as two alternatively spliced isoforms.

REFERENCES

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2. Georgopoulos, C.P., et al. 1980. Identification of the *E. coli* DnaJ gene product. *Mol. Gen. Genet.* 178: 583-588.
3. Suh, W.C., et al. 1998. Interaction of the HSP 70 molecular chaperone, DnaK, with its cochaperone DnaJ. *Proc. Natl. Acad. Sci. USA* 95: 15223-15228.
4. Tomoyasu, T., et al. 1998. Levels of DnaK and DnaJ provide tight control of heat shock gene expression and protein repair in *Escherichia coli*. *Mol. Microbiol.* 30: 567-581.
5. Stewart, G.R., et al. 2004. Analysis of the function of mycobacterial DnaJ proteins by overexpression and microarray profiling. *Tuberculosis* 84: 180-187.
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CHROMOSOMAL LOCATION

Genetic locus: DNAJB14 (human) mapping to 4q23.

PRODUCT

DnaJB14 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 DnaJB14 shRNA Plasmid (h): sc-88854-SH and DnaJB14 shRNA (h) Lentiviral Particles: sc-88854-V as alternate gene silencing products.

For independent verification of DnaJB14 (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-88854A, sc-88854B and sc-88854C.

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

DnaJB14 siRNA (h) is recommended for the inhibition of DnaJB14 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

DnaJB14 (D-6): sc-515383 is recommended as a control antibody for monitoring of DnaJB14 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 DnaJB14 gene expression knockdown using RT-PCR Primer: DnaJB14 (h)-PR: sc-88854-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.