SANTA CRUZ BIOTECHNOLOGY, INC.

HSP 60 siRNA (m): sc-35604

BACKGROUND

The heat shock proteins (HSPs) comprise a group of highly conserved, abundantly expressed proteins with diverse functions, including the assembly and sequestering of multiprotein complexes, transportation of nascent poly-peptide chains across cellular membranes, and the regulation of protein folding. HSPs (also known as molecular chaperones) fall into six general families: HSP 90, HSP 70, HSP 60, the low molecular weight HSPs, the immunophilins, and the HSP 110 family. The constitutively expressed mitochondrial protein HSP 60 shares the ability to recognize and stabilize proteins during folding, assembly and disassembly with other HSP family members. The mitochondrial and cytosolic localization of HSP 60, combined with its binding and catalysis of folding of newly synthesized proteins destined for the mitochondrial matrix, classify this protein as a molecular chaperone. An additional role of HSP 60 is to act as a cell surface marker for γ/δ T cell recognition.

REFERENCES

1. Ritossa, F. 1962. A new puffing pattern induced by temperature shock and DNP in *Drosophila*. Experientia 18: 571-573.
2. Lemeaux, P.G., et al. 1978. Transient rates of synthesis of individual polypeptides in *E. coli* following temperature shifts. Cell 13: 427-434.
3. Kelley, P., et al. 1978. The effect of amino acid analogues and heat shock on gene expression in chicken embryo fibroblasts. Cell 15: 1277-1286.
4. Schlesinger, M.J., et al. 1982. Heat Shock: from Bacteria to Man. Cold Spring Harbor, NY: Cold Spring Harbor Laboratory.
5. Martin, J., et al. 1992. Prevention of protein denaturation under heat stress by the chaperonin HSP 60. Science 258: 995-998.

CHROMOSOMAL LOCATION

Genetic locus: Hspd1 (mouse) mapping to 1 C1.2.

PRODUCT

HSP 60 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 HSP 60 shRNA Plasmid (m): sc-35604-SH and HSP 60 shRNA (m) Lentiviral Particles: sc-35604-V as alternate gene silencing products.

For independent verification of HSP 60 (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-35604A, sc-35604B and sc-35604C.

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

HSP 60 siRNA (m) is recommended for the inhibition of HSP 60 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

HSP 60 (LK1): sc-59567 is recommended as a control antibody for monitoring of HSP 60 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 HSP 60 gene expression knockdown using RT-PCR Primer: HSP 60 (m)-PR: sc-35604-PR (20 µl, 470 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Barone, R., et al. 2016. Skeletal muscle heat shock protein 60 increases after endurance training and induces peroxisome proliferator-activated receptor γ coactivator 1 α1 expression. Sci. Rep. 6: 19781.

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.