SANTA CRUZ BIOTECHNOLOGY, INC.

HSP 90β siRNA (h): sc-35606

BACKGROUND

Heat shock protein (HSP) molecular chaperones are environmental stress-inducible gene products. The human HSP 90 family includes 17 genes that fall into 4 classes: HSP90AA, HSP90AB, HSP90B and TRAP. HSP 90 family members guide the normal folding, intracellular disposition and proteolytic turnover of many key regulators of cell growth, differentiation and survival. HSP 90α, also designated HSP 90A, HSP 86 and LPS-associated protein 2 (LAP2), is a cytosolic enhancer of inducible nitric-oxide synthase (iNOS), with chaperone activity that is important for the transcriptional activity of p53. HSP 90β, also designated HSP 90B, HSP 84 and HSPC2, is a cytosolic protein that participates in signaling pathways with PKC ε to protect cells from external damage, particularly in heat shock-mediated events. GRP 94, also known as tumor rejection antigen 1 (TRA1), ECGP and GP96, localizes to the ER, is highly expressed in BGC-823 human gastric carcinoma cells and is upregulated in human endothelial cells in response to hypoxia by HIF-1. TRAP1 (TNF receptor-associated protein 1), also designated HSP 75, is a mitochondrial matrix component that plays a role in the induction of apoptosis in response to reactive oxygen species.

REFERENCES

1. Wu, J.M., et al. 2003. PKC ε is a unique regulator for HSP 90β gene in heat shock response. J. Biol. Chem. 278: 51143-51149.
2. Whitesell, L., et al. 2005. HSP 90 and the chaperoning of cancer. Nat. Rev. Cancer 5: 761-772.
3. Cowen, L.E., et al. 2005. HSP 90 potentiates the rapid evolution of new traits: drug resistance in diverse fungi. Science 309: 2185-2189.

CHROMOSOMAL LOCATION

Genetic locus: HSP90AB1 (human) mapping to 6p21.1.

PRODUCT

HSP 90β 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HSP 90β shRNA Plasmid (h): sc-35606-SH and HSP 90β shRNA (h) Lentiviral Particles: sc-35606-V as alternate gene silencing products.

For independent verification of HSP 90β (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-35606A, sc-35606B and sc-35606C.

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 90β siRNA (h) is recommended for the inhibition of HSP 90β 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 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 90α/β (F-8): sc-13119 is recommended as a control antibody for monitoring of HSP 90β 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 90β gene expression knockdown using RT-PCR Primer: HSP 90β (h)-PR: sc-35606-PR (20 µl, 443 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Asuthkar, S., et al. 2012. Urokinase-type plasminogen activator receptor (uPAR)-mediated regulation of WNT/β-catenin signaling is enhanced in irradiated medulloblastoma cells. J. Biol. Chem. 287: 20576-20589.
2. Peterson, L.B., et al. 2012. The hERG channel is dependent upon the Hsp90α isoform for maturation and trafficking. Mol. Pharm. 9: 1841-1846.
3. Tsou, Y.L., et al. 2013. Heat shock protein 90: role in enterovirus 71 entry and assembly and potential target for therapy. PLoS ONE 8: e77133.

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.