

prefoldin 3 siRNA (h): sc-40873

BACKGROUND

Molecular chaperones are proteins that assist in the correct folding of other proteins in the crowded molecular environment that exists in living cells. Within this class of proteins, a key role is played by chaperonins, multi-subunit toroidal (i.e. doughnut-shaped) assemblies that undergo major ATP-dependent conformational changes as part of the mechanism of facilitated folding. Prefoldin is a heterohexameric chaperone protein which has the ability to capture unfolded Actin. Six prefoldin polypeptides, prefoldin 1–6, have been identified. Prefoldin 1 is a 122 amino acid protein that binds specifically to cytosolic chaperonin (c-cpn) and transfers target proteins to it. Prefoldin 3 (VBP1 or VHL binding protein-1) forms complexes with VHL and is translocated from perinuclear granules to the nucleus or cytoplasm. Prefoldin 4 is a possible transcription factor. Prefoldin 5 (c-Myc-binding protein Mm-1, Myc modulator 1 or MM-1) is a c-Myc binding protein.

REFERENCES

1. Tsuchiya, H., Iseda, T. and Hino, O. 1996. Identification of a novel protein (VBP-1) binding to the von Hippel-Lindau (VHL) tumor suppressor gene product. *Cancer Res.* 56: 2881-2885.
2. Brinke, A., Green, P.M. and Giannelli, F. 1997. Characterization of the gene (VBP1) and transcript for the von Hippel-Lindau binding protein and isolation of the highly conserved murine homologue. *Genomics* 45: 105-112.
3. Vainberg, I.E., Lewis, S.A., Rommelaere, H., Ampe, C., Vandekerckhove, J., Klein, H.L. and Cowan, N.J. 1998. Prefoldin, a chaperone that delivers unfolded proteins to cytosolic chaperonin. *Cell* 93: 863-873.
4. LocusLink Report (LocusID: 300133). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: VBP1 (human) mapping to Xq28.

PRODUCT

prefoldin 3 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 prefoldin 3 shRNA Plasmid (h): sc-40873-SH and prefoldin 3 shRNA (h) Lentiviral Particles: sc-40873-V as alternate gene silencing products.

For independent verification of prefoldin 3 (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-40873A, sc-40873B and sc-40873C.

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

prefoldin 3 siRNA (h) is recommended for the inhibition of prefoldin 3 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

prefoldin 3 (D-5): sc-390524 is recommended as a control antibody for monitoring of prefoldin 3 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 prefoldin 3 gene expression knockdown using RT-PCR Primer: prefoldin 3 (h)-PR: sc-40873-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Chesnel, F., Couturier, A., Alusse, A., Gagné, J.P., Poirier, G.G., Jean, D., Boisvert, F.M., Hascoet, P., Paillard, L., Arlot-Bonnemains, Y. and Le Goff, X. 2020. The prefoldin complex stabilizes the von Hippel-Lindau protein against aggregation and degradation. *PLoS Genet.* 16: e1009183.

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.