

Proteasemblin siRNA (m): sc-62891

BACKGROUND

Proteasemblin, also known as POMP (proteasome maturation protein), UMP1 or voltage-gated potassium channel β subunit 4.1, is an endoplasmic reticulum (ER) associated protein that functions as a molecular chaperone required for proteasome and immunoproteasome assembly. Essential for cell viability and induced by IFN- γ , Proteasemblin associates with preproteasomes and specifically binds to Proteasome 20S β 1i, β 1, β 5, β 6 and β 7 subunits. Proteasemblin is responsible for mediating the binding of the 20S preproteasome to the ER membrane and is required for incorporation of the β subunits into the 20S proteasome. Proteasemblin is the human homolog of the yeast Ump1 protein. Unlike Ump1, which becomes incorporated into the proteasome, Proteasemblin is degraded upon maturation of the newly formed proteasome.

REFERENCES

1. Griffin, T.A., et al. 2000. Identification of proteasemblin, a mammalian homologue of the yeast protein, Ump1p, that is required for normal proteasome assembly. *Mol. Cell Biol. Res. Commun.* 3: 212-217.
2. Meiners, S., et al. 2003. Inhibition of proteasome activity induces concerted expression of proteasome genes and *de novo* formation of mammalian proteasomes. *J. Biol. Chem.* 278: 21517-21525.
3. Jayarapu, K. and Griffin, T.A. 2004. Protein-protein interactions among human 20S proteasome subunits and proteasemblin. *Biochem. Biophys. Res. Commun.* 314: 523-528.
4. Chen, Q., et al. 2005. RNA interference toward UMP1 induces proteasome inhibition in *Saccharomyces cerevisiae*: evidence for protein oxidation and autophagic cell death. *Free Radic. Biol. Med.* 38: 226-234.
5. Heink, S., et al. 2005. IFN- γ -induced immune adaptation of the proteasome system is an accelerated and transient response. *Proc. Natl. Acad. Sci. USA* 102: 9241-9246.
6. Hirano, Y., et al. 2005. A heterodimeric complex that promotes the assembly of mammalian 20S proteasomes. *Nature* 437: 1381-1385.

CHROMOSOMAL LOCATION

Genetic locus: Pomp (mouse) mapping to 5 G3.

PRODUCT

Proteasemblin 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Proteasemblin shRNA Plasmid (m): sc-62891-SH and Proteasemblin shRNA (m) Lentiviral Particles: sc-62891-V as alternate gene silencing products.

For independent verification of Proteasemblin (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-62891A, sc-62891B and sc-62891C.

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

Proteasemblin siRNA (m) is recommended for the inhibition of Proteasemblin 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 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

Proteasemblin (B-1): sc-393267 is recommended as a control antibody for monitoring of Proteasemblin 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 Proteasemblin gene expression knockdown using RT-PCR Primer: Proteasemblin (m)-PR: sc-62891-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.