

PSMC3 siRNA (m): sc-76276

BACKGROUND

In eukaryotic cells, selective breakdown of cellular proteins is ensured by their ubiquitination and subsequent degradation by the 26S Proteasome. The 26S Proteasome is a protease complex that selectively breaks down proteins that have been modified by polyubiquitin chains. It is made up of two multi-subunit complexes: the 20S Proteasome chamber, which serves as the proteolytic core of the complex, and two 19S regulatory particles which recognize and unfold ubiquitinated proteins. PSMC3 (Proteasome 26S subunit ATPase 3), also known as TBP1 (Tat-binding protein 1), is a 439 amino acid member of the AAA ATPase family. Localized to both the nucleus and the cytoplasm, PSMC3 functions as a subunit of the 19S regulatory complex and is involved in regulating the substrate specificity of the 26S Proteasome. Additionally, PSMC3 interacts with the HIV protein HIV-1 Tat and, via this interaction, mediates the association of the viral protein with transcription complexes.

REFERENCES

1. Hoyle, J., et al. 1997. Localization of genes encoding two human one-domain members of the AAA family: PSMC5 (the thyroid hormone receptor-interacting protein, TRIP1) and PSMC3 (the Tat-binding protein, TBP1). *Hum. Genet.* 99: 285-288.
2. Tanahashi, N., et al. 1998. Chromosomal localization and immunological analysis of a family of human 26S proteasomal ATPases. *Biochem. Biophys. Res. Commun.* 243: 229-232.
3. Conticello, S.G., et al. 2003. The Vif protein of HIV triggers degradation of the human antiretroviral DNA deaminase APOBEC3G. *Curr. Biol.* 13: 2009-2013.
4. Apcher, G.S., et al. 2003. Human immunodeficiency virus-1 Tat protein interacts with distinct proteasomal α and β subunits. *FEBS Lett.* 553: 200-204.
5. Shindo, K., et al. 2003. The enzymatic activity of CEM15/Apobec-3G is essential for the regulation of the infectivity of HIV-1 virion but not a sole determinant of its antiviral activity. *J. Biol. Chem.* 278: 44412-44416.

CHROMOSOMAL LOCATION

Genetic locus: Psmc3 (mouse) mapping to 2 E1.

PRODUCT

PSMC3 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 PSMC3 shRNA Plasmid (m): sc-76276-SH and PSMC3 shRNA (m) Lentiviral Particles: sc-76276-V as alternate gene silencing products.

For independent verification of PSMC3 (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-76276A, sc-76276B and sc-76276C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

PSMC3 siRNA (m) is recommended for the inhibition of PSMC3 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

PSMC3 (28-K): sc-100462 is recommended as a control antibody for monitoring of PSMC3 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 PSMC3 gene expression knockdown using RT-PCR Primer: PSMC3 (m)-PR: sc-76276-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.