



PA200 siRNA (h): sc-94428

BACKGROUND

PA200 (proteasome activator 200 kDa), also known as PSME4 (proteasome (prosome, macropain) activator subunit 4), is a 1,843 amino acid nuclear protein that contains six HEAT (huntington, elongation factor 3, PR65/A, TOR) repeats, which are conserved residues that form the hydrophobic domain core and are usually found in proteins that are involved in intracellular transport. Existing as a homodimer, PA200 interacts with the 20S and 26S Proteasomes and activates proteasomal cleavage of peptides in an energy-independent manner. PA200 and proteasomes function together within cells and respond to specific radiation-induced damage independent of the stage of cell cycle arrest. Broadly expressed, PA200 may also be involved in spermatogenesis and in DNA repair double-strand breaks (DSBs). Four isoforms of PA200 exist due to alternative splicing events.

REFERENCES

1. Ustrell, V., et al. 2002. PA200, a nuclear proteasome activator involved in DNA repair. *EMBO J.* 21: 3516-3525.
2. Kajava, A.V., et al. 2004. New HEAT-like repeat motifs in proteins regulating proteasome structure and function. *J. Struct. Biol.* 146: 425-430.
3. Ortega, J., et al. 2005. The axial channel of the 20S Proteasome opens upon binding of the PA200 activator. *J. Mol. Biol.* 346: 1221-1227.
4. Ustrell, V., et al. 2005. Purification and assay of proteasome activator PA200. *Meth. Enzymol.* 398: 321-329.
5. Gomes, A.V., et al. 2006. Mapping the murine cardiac 26S Proteasome complexes. *Circ. Res.* 99: 362-371.
6. McCulloch, S., et al. 2006. Bim3-1 is an allele of UBP3, a ubiquitin protease that appears to act during transcription of damaged DNA. *J. Mol. Biol.* 363: 660-672.
7. Khor, B., et al. 2006. Proteasome activator PA200 is required for normal spermatogenesis. *Mol. Cell. Biol.* 26: 2999-3007.
8. Blickwedehl, J., et al. 2007. Proteasomes and proteasome activator 200 kDa (PA200) accumulate on chromatin in response to ionizing radiation. *Radiat. Res.* 167: 663-674.

CHROMOSOMAL LOCATION

Genetic locus: PSME4 (human) mapping to 2p16.2.

PRODUCT

PA200 siRNA (h) is a pool of 2 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 PA200 shRNA Plasmid (h): sc-94428-SH and PA200 shRNA (h) Lentiviral Particles: sc-94428-V as alternate gene silencing products.

For independent verification of PA200 (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-94428A and sc-94428B.

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

PA200 siRNA (h) is recommended for the inhibition of PA200 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 contains 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PA200 gene expression knockdown using RT-PCR Primer: PA200 (h)-PR: sc-94428-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.