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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic)

S-100A10 shRNA (h) Lentiviral Particles: sc-44084-V

BACKGROUND

The S-100 family of calcium-activated proteins interact with a range of target proteins to modulate biological signaling pathways. Numerous cancer cell lines overexpress the plasminogen receptor S-100A10 on the extracellular cell surface, where it forms a heterotetrameric complex with Annexin II, though this association is not required for plasma membrane localization or binding and activation of plasminogen. Additionally, S-100A10 acts as a cellular chaperone for hepatitis B (Hep B) virus polymerase. Hep B virus polymerase normally localizes to the cytoplasm only, though in the presence of S-100A10 a portion relocates to the nucleus, implying a role for S-100A10 and intracellular calcium in the process of viral replication.

REFERENCES

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- Ruse, M., et al. 2001. S-100A7, S-100A10, and S-100A11 are transglutaminase substrates. *Biochemistry* 40: 3167-3173.
- El-Rifai, W., et al. 2002. Gastric cancers overexpress S-100A calcium-binding proteins. *Cancer Res.* 62: 6823-6826.
- Broome, A.M., et al. 2003. S-100 protein subcellular localization during epidermal differentiation and psoriasis. *J. Histochem. Cytochem.* 51: 675-685.
- Choi, J., et al. 2003. Association of hepatitis B virus polymerase with promyelocytic leukemia nuclear bodies mediated by the S-100 family protein p11. *Biochem. Biophys. Res. Commun.* 305: 1049-1056.
- Daub, B., et al. 2003. Expression of members of the S-100 Ca²⁺-binding protein family in guinea-pig smooth muscle. *Cell Calcium* 33: 1-10.

CHROMOSOMAL LOCATION

Genetic locus: S100A10 (human) mapping to 1q21.3.

PRODUCT

S-100A10 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see S-100A10 siRNA (h): sc-44084 and S-100A10 shRNA Plasmid (h): sc-44084-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80°C . Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4°C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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

APPLICATIONS

S-100A10 shRNA (h) Lentiviral Particles is recommended for the inhibition of S-100A10 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 lentiviral transducing particles per milliliter; contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

S-100A10 (H-80): sc-50450 is recommended as a control antibody for monitoring of S-100A10 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor S-100A10 gene expression knockdown using RT-PCR Primer: S-100A10 (h)-PR: sc-44084-PR (20 μ l, 434 bp). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.