SANTA CRUZ BIOTECHNOLOGY, INC.

PU.1 siRNA (h): sc-36330

BACKGROUND

The Ets transcription factor family (Ets-1, Ets-2, Erg-1–3, Elk-1, Elf-1, Elf-5, NERF, PU.1, PEA3, ERM, FEV, ER81, Fli-1, TEL, Spi-B, ESE-1, ESE-3A, Net, ABT1 and ERF) are DNA-binding proteins that influence lymphoid development and activity. The Ets family monomeric proteins bind the consensus DNA site GGA(A/T) through a unique winged helix-turn-helix motif known as the Ets domain. PU.1 (Spi-1/Spi-A), Spi-B and Spi-C are closely related Ets family members which share a conserved divergent sequence within the Ets domain that enables their binding to the non-canonical AGAA sites. PU.1 transactivates a large number of B cell genes, such as those encoding CD72, CD20 and Btk, and Spi-B enhances expression of many of these same target genes. PU.1 is expressed in a wide variety of hematopoetic cells, including B cells, early T-cells, megakaryocytes, granulocytes, mast cells, immature erythrocytes and myeloid cells. Alternatively, Spi-B expression is limited to B cells and immature T cells, where expression accumulates through T-lineage commitment and then is dramatically absent following the β-selection checkpoint.

REFERENCES

1. Kola, I., et al. 1993. The Ets1 transcription factor is widely expressed during murine embryo development and is associated with mesodermal cells involved in morphogenetic processes such as organ formation. Proc. Natl. Acad. Sci. USA 90: 7588-7592.
2. Chen, H., et al. 1995. PU.1 (Spi-1) autoregulates its expression in myeloid cells. Oncogene 11: 1549-1560.
3. Chen, H.M., et al. 1995. Neutrophils and monocytes express high levels of PU.1 (Spi-1) but not Spi-B. Blood 85: 2918-2928.

CHROMOSOMAL LOCATION

Genetic locus: SPI1 (human) mapping to 11p11.2.

PRODUCT

PU.1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PU.1 shRNA Plasmid (h): sc-36330-SH and PU.1 shRNA (h) Lentiviral Particles: sc-36330-V as alternate gene silencing products.

For independent verification of PU.1 (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-36330A, sc-36330B and sc-36330C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PU.1 siRNA (h) is recommended for the inhibition of PU.1 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 µM in 66 µl. 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

PU.1 (C-3): sc-390405 is recommended as a control antibody for monitoring of PU.1 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PU.1 gene expression knockdown using RT-PCR Primer: PU.1 (h)-PR: sc-36330-PR (20 µl, 518 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Domínguez-Soto, A., et al. 2005. PU.1 regulates the tissue-specific expression of dendritic cell-specific intercellular adhesion molecule (ICAM)-3-grabbing nonintegrin. J. Biol. Chem. 280: 33123-33131.
2. Sarma, N.J., et al. 2014. Hepatitis C virus-induced changes in microRNA 107 (miRNA-107) and miRNA-449a modulate CCL2 by targeting the interleukin-6 receptor complex in hepatitis. J. Virol. 88: 3733-3743.
3. Grassilli, S., et al. 2016. A network including PU.1, Vav1 and miR-142-3p sustains ATRA-induced differentiation of acute promyelocytic leukemia cells—a short report. Cell. Oncol. 39: 483-489.
4. Ewelina, W., et al. 2017. Downregulation of PARP1 transcription by promoter-associated E2F4-RBL2-HDAC1-BRM complex contributes to repression of pluripotency stem cell factors in human monocytes. Sci. Rep. 7: 9483.
5. Vezzali, F., et al. 2018. Vav1 is necessary for PU.1 mediated upmodulation of miR-29b in acute myeloid leukaemia-derived cells. J. Cell. Mol. Med. 22: 3149-3158.
6. Samaniego, R., et al. 2020. Folate receptor β (FRβ) expression in tissue-resident and tumor-associated macrophages associates with and depends on the expression of PU.1. Cells 9: 1445.

RESEARCH USE

For research use only, not for use in diagnostic procedures.