SANTA CRUZ BIOTECHNOLOGY, INC.

Spi-C siRNA (h): sc-76561

BACKGROUND

The Ets transcription factor family is comprised of DNA-binding proteins that influence lymphoid development and activity and bind the consensus DNA site GGA(A/T) through a unique winged helix-turn-helix motif known as the Ets domain. Spi-B and Spi-C (also known as SPIC) are closely related Ets family members which share a conserved divergent sequence within the Ets domain that enables their binding to non-canonical AGAA sites. Spi-C is a 248 amino acid protein that localizes to the nucleus and, like other Ets family members, binds DNA as a monomer and plays a role in transcriptional regulation. Additionally, Spi-C is thought to control the development of red pulp macrophages, thereby contributing to iron homeostasis and red blood cell recycling. Human Spi-C shares 65% amino acid identity with its mouse counterpart, suggesting a conserved role between species.

REFERENCES

1. Carlsson, R., et al. 2002. Genomic structure of mouse Spi-C and genomic structure and expression pattern of human Spi-C. Gene 299: 271-278.
2. Kageyama, S., et al. 2006. The role of ETS transcription factors in transcription and development of mouse preimplantation embryos. Biochem. Biophys. Res. Commun. 344: 675-679.
3. Carlsson, R., et al. 2006. Spi-C and STAT6 can cooperate to stimulate IgE germline transcription. Biochem. Biophys. Res. Commun. 344: 1155-1160.
4. Guillouf, C., et al. 2006. Spi-1/PU.1 oncoprotein affects splicing decisions in a promoter binding-dependent manner. J. Biol. Chem. 281: 19145-19155.
5. Schweitzer, B.L., et al. 2006. Spi-C has opposing effects to PU.1 on gene expression in progenitor B cells. J. Immunol. 177: 2195-2207.
6. Zhu, X., et al. 2008. Transgenic expression of Spi-C impairs B-cell development and function by affecting genes associated with Bcr signaling. Eur. J. Immunol. 38: 2587-2599.
7. Uchiya, K. and Nikai, T. 2008. Salmonella virulence factor Spi-C is involved in expression of flagellin protein and mediates activation of the signal transduction pathways in macrophages. Microbiology 154: 3491-3502.
8. Kohyama, M., et al. 2009. Role for Spi-C in the development of red pulp macrophages and splenic iron homeostasis. Nature 457: 318-321.

CHROMOSOMAL LOCATION

Genetic locus: SPIC (human) mapping to 12q23.2.

PRODUCT

Spi-C 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 Spi-C shRNA Plasmid (h): sc-76561-SH and Spi-C shRNA (h) Lentiviral Particles: sc-76561-V as alternate gene silencing products.

For independent verification of Spi-C (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-76561A, sc-76561B and sc-76561C.

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

Spi-C siRNA (h) is recommended for the inhibition of Spi-C 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

Spi-C (C-2): sc-514526 is recommended as a control antibody for monitoring of Spi-C 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Spi-C gene expression knockdown using RT-PCR Primer: Spi-C (h)-PR: sc-76561-PR (20 µl). 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.