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SANTA CRUZ BIOTECHNOLOGY, INC.

LITAF siRNA (m): sc-45685

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

Lipopolysaccharide (LPS) is a potent stimulator of monocytes and macrophages, causing secretion of tumor necrosis factor α (TNFα) and other inflammatory mediators. The inflammatory response to bacteria and bacterial products, such as LPS, is mediated by a variety of secreted factors, but cytotoxic effects of LPS have been ascribed to the TNFα activity. LITAF (LPS-induced TNFα factor), Stat6B, and the LITAF-Stat6B complex all play a role in the regulation of inflammatory cytokines in response to LPS or p53 stimulation in mammalian cells. LITAF is a nuclear protein crucial in TNFα gene transcription regulation. High levels of expression of LITAF mRNA have been observed predominantly in the placenta, peripheral blood leukocytes, lymph nodes and the spleen.

REFERENCES

1. Myokai, F., Takashiba, S., Lebo, R. and Amar, S.I. 1999. A novel lipopolysaccharide-induced transcription factor regulating tumor necrosis factor-α gene expression: molecular cloning, sequencing, characterization and chromosomal assignment. Proc. Natl. Acad. Sci. USA 96: 4518-4523.
2. Zhou, H.R., Islam, Z. and Pestka, J.J. 2003. Kinetics of lipopolysaccharide-induced transcription factor activation/inactivation and relation to proinflammatory gene expression in the murine spleen. Toxicol. Appl. Pharmacol. 187: 147-161.
3. Matsumura, Y., Nishigori, C., Horio, T. and Miyachi, Y. 2004. PIG7/LITAF gene mutation and overexpression of its gene product in extramammary Paget's disease. Int. J. Cancer 111: 218-223.
4. Bolcato-Bellemin, A.L., Mattei, M.G., Fenton, M. and Amar, S. 2004. Molecular cloning and characterization of mouse LITAF cDNA: role in the regulation of tumor necrosis factor-α (TNFα) gene expression. J. Endotoxin Res. 10: 15-23.
5. Tang, X., Marciano, D.L., Leeman, S.E. and Amar, S.I. 2005. LPS induces the interaction of a transcription factor, LPS-induced TNFα factor, and Stat6B with effects on multiple cytokines. Proc. Natl. Acad. Sci. USA 102: 5132-5137.

CHROMOSOMAL LOCATION

Genetic locus: Litaf (mouse) mapping to 16 A1.

PRODUCT

LITAF 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see LITAF shRNA Plasmid (m): sc-45685-SH and LITAF shRNA (m) Lentiviral Particles: sc-45685-V as alternate gene silencing products.

For independent verification of LITAF (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-45685A, sc-45685B and sc-45685C.

RESEARCH USE

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

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

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

LITAF (C-5): sc-166719 is recommended as a control antibody for monitoring of LITAF 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 LITAF gene expression knockdown using RT-PCR Primer: LITAF (m)-PR: sc-45685-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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