SANTA CRUZ BIOTECHNOLOGY, INC.

GM-CSF siRNA (m): sc-39392

BACKGROUND

Colony stimulating factors (CSFs) were initially characterized by their ability to stimulate *in vitro* colony formation by hematopoietic progenitor cells in semisolid media. Several of these CSFs have been assigned an interleukin number, while three (GM-CSF, G-CSF and M-CSF) have retained their CSF designations. The human granulocyte-macrophage colony stimulating factor (GM-CSF) is a pleiotropic cytokine with a 17 amino acid signal peptide that is cleaved to produce the mature form of 127 amino acids. The mature murine GM-CSF protein is 124 amino acids and shares 60% homology with the human GM-CSF protein. GM-CSF is a glycoprotein that can stimulate the proliferation of hematopoietic cells including granulocytes and macrophages. It has been shown to promote the phosphorylation of $cPLA_2$ in human neutrophils. The phosphorylation of $cPLA_2$ was accompanied by an increase in the enzyme activity.

REFERENCES

1. Suda, T., et al. 1990. Identification of a novel thymocyte growth-promoting factor derived from B cell lymphomas. Cell. Immunol. 129: 228-240.
2. Nozaki, S., et al. 1991. Augmentation of granulocyte/macrophage colony-stimulating factor expression by ultraviolet irradiation is mediated by interleukin 1 in Pam 212 keratinocytes. J. Invest. Dermatol. 97: 10-14.
3. Moore, M.A. 1991. The clinical use of colony stimulating factors. Annu. Rev. Immunol. 9: 159-191.
4. Abrams, J.S., et al. 1992. Strategies of anti-cytokine monoclonal antibody development: immunoassay of IL-10 and IL-5 in clinical samples. Immunol. Rev. 127: 5-24.
5. Freund, M. and Kleine, H.D. 1993. The role of GM-CSF in infection. Infection 20: S84-S92.
6. Costello, R.T. 1993. Therapeutic use of granulocyte-macrophage colony-stimulating factor (GM-CSF). A review of recent experience. Acta Oncol. 32: 403-408.
7. Sander, B., et al. 1994. Similar frequencies and blood and spleen. Cytokine detection by immunoassay and intracellular immunostaining. J. Immunol. Methods 166: 201-214.

CHROMOSOMAL LOCATION

Genetic locus: Csf2 (mouse) mapping to 11 B1.3.

PRODUCT

GM-CSF 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 GM-CSF shRNA Plasmid (m): sc-39392-SH and GM-CSF shRNA (m) Lentiviral Particles: sc-39392-V as alternate gene silencing products.

For independent verification of GM-CSF (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-39392A, sc-39392B and sc-39392C.

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

GM-CASF siRNA (m) is recommended for the inhibition of GM-CASF 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

GM-CSF (B6-2-hGMCSF): sc-32753 is recommended as a control antibody for monitoring of GM-CSF 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 GM-CSF gene expression knockdown using RT-PCR Primer: GM-CSF (m)-PR: sc-39392-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.