

FMIP siRNA (m): sc-145205

BACKGROUND

Although the macrophage colony stimulating factor (M-CSF) and its receptor, c-Fms, are involved in the survival and proliferation of hematopoietic cells, little is known about the signalling events leading to differentiation into mature blood cells. An Fms-interacting protein, FMIP, transiently binds to M-CSF-activated Fms-molecules. This binding results in a rapid phosphorylation of FMIP within its Fms-binding domain, thereby dissociating Fms and FMIP. Endogenous levels of FMIP may form a threshold that decide whether bipotential progenitor cells differentiate into macrophages or granulocytes. Myeloid progenitor cells express low levels of endogenous FMIP and, upon M-CSF specific signalling, are differentiated into macrophages. Overexpression of FMIP may saturate Fms, which results in predominant cytoplasmic expression of FMIP and favors granulocyte differentiation.

REFERENCES

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2. Gliniak, S.C. and Rohrschneider, R.L. 1990. Expression of the M-CSF receptor is controlled posttranscriptionally by the dominant actions of GM-CSF or multi-CSF. *Cell* 63: 1073-1083.
3. Cross, M.A., Heyworth, C.M. and Dexter, T.M. 1997. The Molecular Basis of Cellular Defence Mechanisms, Ciba Foundation Symposium. New York: John Wiley & Sons, 3-16.
4. Broudy, V.C. 1997. Stem cell factor and hematopoiesis. *Blood* 90: 1345-1364.
5. Tamura, T., Mancini, A., Joos, H., Koch, A., Hakim, C., Dumanski, J., Weidner, K.M. and Neimann, H. 1999. FMIP, a novel Fms-interacting protein, affects granulocyte/macrophage differentiation. *Oncogene* 18: 6488-6495.

CHROMOSOMAL LOCATION

Genetic locus: Thoc5 (mouse) mapping to 11 A1.

PRODUCT

FMIP 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FMIP shRNA Plasmid (m): sc-145205-SH and FMIP shRNA (m) Lentiviral Particles: sc-145205-V as alternate gene silencing products.

For independent verification of FMIP (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-145205A, sc-145205B and sc-145205C.

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.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

FMIP siRNA (m) is recommended for the inhibition of FMIP 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 μ M in 66 μ l. 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

FMIP (H-8): sc-514146 is recommended as a control antibody for monitoring of FMIP 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 FMIP gene expression knockdown using RT-PCR Primer: FMIP (m)-PR: sc-145205-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.