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Mig-6 siRNA (m): sc-45705

BACKGROUND

Mitogen-inducible gene 6 protein (Mig-6), also designated Gene 33 or RALT, belongs to the Mig-6 family. The gene encoding for Mig-6 maps to chromosome 1p36.23. Mig-6 is a cytoplasmic protein acting as a feedback inhibitor of ErbB-2 mitogenic function and can suppress ErbB-2 oncogenic activity. The expression of Mig-6 is upregulated with cell growth. Mig-6 binds to the epidermal growth factor receptor (EGFR) upon EGF stimulation and is considered a negative feedback regulator of EGFR and a potential tumor suppressor. Mig-6 induces transcriptional activation of NF κ B by binding to its inhibitor, I κ B- α . It enables the cell to respond persistently to chronic stress. Mig-6 mRNA levels increase in response to stress such as diabetic nephropathy, vasoactive peptides or mechanical strain. Mig-6 is expressed in liver, placenta and lung.

REFERENCES

- Wick, M., et al. 1995. Identification of a novel mitogen-inducible gene (Mig-6): regulation during G₁ progression and differentiation. *Exp. Cell Res.* 219: 527-535.
- Makkinje, A., et al. 2000. Gene 33/Mig-6, a transcriptionally inducible adapter protein that binds GTP-Cdc42 and activates SAPK/JNK. A potential marker transcript for chronic pathologic conditions, such as diabetic nephropathy. Possible role in the response to persistent stress. *J. Biol. Chem.* 275: 17838-17847.
- Fiorentino, L., et al. 2000. Inhibition of ErbB-2 mitogenic and transforming activity by RALT, a mitogen-induced signal transducer which binds to the ErbB-2 kinase domain. *Mol. Cell. Biol.* 20: 7735-7750.
- Hackel, P.O., et al. 2001. Mig-6 is a negative regulator of the epidermal growth factor receptor signal. *Biol. Chem.* 382: 1649-1662.

CHROMOSOMAL LOCATION

Genetic locus: Errf1 (mouse) mapping to 4 E2.

PRODUCT

Mig-6 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 Mig-6 shRNA Plasmid (m): sc-45705-SH and Mig-6 shRNA (m) Lentiviral Particles: sc-45705-V as alternate gene silencing products.

For independent verification of Mig-6 (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-45705A, sc-45705B and sc-45705C.

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

Mig-6 shRNA Plasmid (m) is recommended for the inhibition of Mig-6 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

Mig-6 (D-4): sc-390905 is recommended as a control antibody for monitoring of Mig-6 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Mig-6 gene expression knockdown using RT-PCR Primer: Mig-6 (m)-PR: sc-45705-PR (20 μ l, 590 bp). 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 or our catalog for detailed protocols and support products.