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Mig-6 shRNA (m) Lentiviral Particles: sc-45705-V

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 1p36.12-p36.33. 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

1. 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.
2. 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.
3. 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.
4. 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: Erff1 (mouse) mapping to 4 E2.

PRODUCT

Mig-6 shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0 x 10⁶ infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Mig-6 siRNA (m): sc-45705 and Mig-6 shRNA Plasmid (m): sc-45705-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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

APPLICATIONS

Mig-6 shRNA (m) Lentiviral Particles is recommended for the inhibition of Mig-6 expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0 x 10⁶ infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

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.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.