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ERGIC-53 siRNA (h): sc-45246

BACKGROUND

Lectin mannose-binding 1, also designated vesicular integral-membrane protein (VIP36) and lectin mannose-binding 2, also designated ER-Golgi intermediate compartment (ERGIC-53) comprise a family of membrane bound, ubiquitous proteins involved in the selective transport of newly synthesized glycoproteins from the endoplasmic reticulum (ER) to the ER-Golgi intermediate compartment (ERGIC). VIP36 acts as an intracellular lectin in the early secretory pathway. It is involved in the sorting and transport of glycoproteins carrying high mannose-type glycans. ERGIC-53, a mannose-specific lectin, recognizes sugar residues of glycoproteins and glycolipids. It mediates the sorting and recycling of proteins and/or lipids. Null expression of ERGIC-53, also designated LMAN1, results in a rare autosomal recessive bleeding disorder that causes combined deficiency of both coagulation factors V and VIII.

REFERENCES

- Schindler, R., et al. 1993. ERGIC-53, a membrane protein of the ER-Golgi intermediate compartment, carries an ER retention motif. *Eur. J. Cell Biol.* 61: 1-9.
- Kappeler, F., et al. 1994. A dual role for COOH-terminal lysine residues in pre-Golgi retention and endocytosis of ERGIC-53. *J. Biol. Chem.* 269: 6279-6281.
- Hauri, H.P., et al. 2002. Lectins and protein traffic early in the secretory pathway. *Biochem. Soc. Symp.* 69: 73-82.
- Cunningham, M.A., et al. 2003. LMAN1 is a molecular chaperone for the secretion of coagulation factor VIII. *J. Thromb. Haemost.* 1: 2360-2367.

CHROMOSOMAL LOCATION

Genetic locus: LMAN1 (human) mapping to 18q21.32.

PRODUCT

ERGIC-53 siRNA (h) 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 ERGIC-53 shRNA Plasmid (h): sc-45246-SH and ERGIC-53 shRNA (h) Lentiviral Particles: sc-45246-V as alternate gene silencing products.

For independent verification of ERGIC-53 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-45246A, sc-45246B and sc-45246C.

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

ERGIC-53 siRNA (h) is recommended for the inhibition of ERGIC-53 expression in human 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

ERGIC-53 (F-3): sc-398777 is recommended as a control antibody for monitoring of ERGIC-53 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 ERGIC-53 gene expression knockdown using RT-PCR Primer: ERGIC-53 (h)-PR: sc-45246-PR (20 μ l, 513 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Fukamachi, M., et al. 2018. Multiple coagulation factor deficiency protein 2 as a crucial component in metastasis of human oral cancer. *Exp. Cell Res.* 368: 119-125.

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.