

p55 siRNA (h2): sc-156153

BACKGROUND

P55 is an extensively palmitoylated erythrocyte membrane protein, and a member of the MAGUK family. P55 also resists salt extraction, resulting in a high affinity for the plasma membrane. P55 contains a PDZ/DHR domain, a conserved SH-3 domain that appears to suppress tyrosine kinase activity of various oncoproteins, a 39 amino acid motif that binds to cytoskeletal protein 4.1R, and a guanylate kinase-like domain. Interaction with glycophorin C (GPC) and 4.1R suggests that p55 may play a role in the dynamic regulation in the erythrocyte membrane. In addition, p55 gene expression *in vivo* may be associated with a CpG island. p55 is constitutively expressed in K-562 erythroleukemia cells during erythropoiesis and undergoes a two-fold amplification after induction.

REFERENCES

1. Ruff, P., et al. 1991. Molecular identification of a major palmitoylated erythrocyte membrane protein containing the Src homology 3 motif. *Proc. Natl. Acad. Sci. USA* 88: 6595-6599.
2. Das, A.K., et al. 1992. Fatty acylation of a 55 kDa membrane protein of human erythrocytes. *Biochem. Biophys. Acta* 1108: 128-132.
3. Marfatia, S.M., et al. 1995. Identification of the protein 4.1 binding interface on glycophorin C and p55, a homologue of the *Drosophila* discs-large tumor suppressor protein. *J. Biol. Chem.* 270: 715-719.
4. Kim, A.C., et al. 1996. Complete genomic organization of the human erythroid p55 gene (MPP1), a membrane-associated guanylate kinase homologue. *Genomics* 31: 223-229.
5. Nunomura, W., et al. 2000. Regulation of protein 4.1R, p55, and glycophorin C ternary complex in human erythrocyte membrane. *J. Biol. Chem.* 275: 24540-24546.

CHROMOSOMAL LOCATION

Genetic locus: MPP1 (human) mapping to Xq28.

PRODUCT

p55 siRNA (h2) 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 p55 shRNA Plasmid (h2): sc-156153-SH and p55 shRNA (h2) Lentiviral Particles: sc-156153-V as alternate gene silencing products.

For independent verification of p55 (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-156153A, sc-156153B and sc-156153C.

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

p55 siRNA (h2) is recommended for the inhibition of p55 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

p55 (A-7): sc-374506 is recommended as a control antibody for monitoring of p55 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 p55 gene expression knockdown using RT-PCR Primer: p55 (h2)-PR: sc-156153-PR (20 μ l, 569 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.