

PHACTR1 siRNA (h): sc-95456

BACKGROUND

In eukaryotes, the phosphorylation and dephosphorylation of proteins on serine and threonine residues is an essential means of regulating a broad range of cellular functions, including division, homeostasis and apoptosis. A group of proteins that are intimately involved in this process are the protein phosphatases. PHACTR1 (phosphatase and Actin regulator 1), also known as RPEL or RPEL1, is a 580 amino acid cytoplasmic protein belonging to the phosphatase and Actin regulator family. Existing as two alternatively spliced isoforms, PHACTR1 contains RPEL repeats and is encoded by a gene located on human chromosome 6, which contains around 1,200 genes and 170 million base pairs. Deletion of a portion of the q arm of chromosome 6 is associated with early onset intestinal cancer suggesting the presence of a cancer susceptibility locus. Porphyria cutanea tarda is associated with chromosome 6 through the HFE gene which, when mutated, predisposes an individual to developing this porphyria.

REFERENCES

- Allen, P.B., et al. 2004. Phactrs 1-4: a family of protein phosphatase 1 and Actin regulatory proteins. *Proc. Natl. Acad. Sci. USA* 101: 7187-7192.
- Online Mendelian Inheritance in Man, OMIM[™]. 2004. Johns Hopkins University, Baltimore, MD. MIM Number: 608723. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
- Drögemüller, C., et al. 2005. Assignment of the PHACTR1 gene to bovine chromosome 23q24 by fluorescence *in situ* hybridization and radiation hybrid mapping. *Cytogenet. Genome Res.* 109: 533.

CHROMOSOMAL LOCATION

Genetic locus: PHACTR1 (human) mapping to 6p24.1.

PRODUCT

PHACTR1 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 PHACTR1 shRNA Plasmid (h): sc-95456-SH and PHACTR1 shRNA (h) Lentiviral Particles: sc-95456-V as alternate gene silencing products.

For independent verification of PHACTR1 (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-95456A, sc-95456B and sc-95456C.

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

PHACTR1 siRNA (h) is recommended for the inhibition of PHACTR1 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

PHACTR1 (E-2): sc-514800 is recommended as a control antibody for monitoring of PHACTR1 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 λ BP-HRP: sc-516132 or m-IgG λ BP-HRP (Cruz Marker): sc-516132-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 λ BP-FITC: sc-516185 or m-IgG λ BP-PE: sc-516186 (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 PHACTR1 gene expression knockdown using RT-PCR Primer: PHACTR1 (h)-PR: sc-95456-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Codina-Fauteux, V.A., et al. 2018. PHACTR1 splicing isoforms and eQTLs in atherosclerosis-relevant human cells. *BMC Med. Genet.* 19: 97.
- Yang, Y., et al. 2020. PHACTR1 is associated with disease progression in Chinese Moyamoya disease. *PeerJ* 8: e8841.

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.