

VHL siRNA (m): sc-36817

BACKGROUND

Individuals harboring germline mutations in the tumor suppressor gene von Hippel-Lindau (VHL) exhibit an increased susceptibility to a variety of tumors including renal carcinoma, hemangioblastoma of the central nervous system and pheochromocytoma. The Elongin (SIII) complex has been identified as the functional target of the VHL protein. Elongin (SIII) is a heterotrimer composed of a transcriptional active subunit designated Elongin A and two regulatory subunits designated Elongin B and Elongin C. VHL functions by binding to the Elongin B and C subunits, inhibiting the transcriptional efficacy of the Elongin (SIII) complex. Different isoforms of VHL have been observed, encoded by alternatively spliced transcript variants. The molecular weight of each isoform varies between species.

REFERENCES

1. Seizinger, B.R., et al. 1988. Von Hippel-Lindau disease maps to the region of chromosome 3 associated with renal cell carcinoma. *Nature* 332: 268-269.
2. Decker, H.J., et al. 1989. Loss of heterozygosity on 3p in a renal cell carcinoma in von Hippel-Lindau syndrome. *Cancer Genet. Cytogenet.* 39: 289-293.
3. Tory, K., et al. 1989. Specific genetic change in tumors associated with von Hippel-Lindau disease. *J. Natl. Cancer Inst.* 81: 1097-1101.
4. Garrett, K.P., et al. 1994. Molecular cloning of an essential subunit of RNA polymerase II elongation factor SIII. *Proc. Natl. Acad. Sci. USA* 91: 5237-5241.
5. Krumm, A., et al. 1995. Tumor suppression and transcription elongation: the dire consequences of changing partners. *Science* 269: 1400-1401.

CHROMOSOMAL LOCATION

Genetic locus: Vhl (mouse) mapping to 6 E3.

PRODUCT

VHL 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 VHL shRNA Plasmid (m): sc-36817-SH and VHL shRNA (m) Lentiviral Particles: sc-36817-V as alternate gene silencing products.

For independent verification of VHL (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-36817A, sc-36817B and sc-36817C.

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

VHL siRNA (m) is recommended for the inhibition of VHL 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

VHL (D-7): sc-55506 is recommended as a control antibody for monitoring of VHL 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 VHL gene expression knockdown using RT-PCR Primer: VHL (m)-PR: sc-36817-PR (20 μ l, 440 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Li, D., et al. 2017. Hepatic hypoxia-inducible factors inhibit PPAR α expression to exacerbate acetaminophen induced oxidative stress and hepatotoxicity. *Free Radic. Biol. Med.* 110: 102-116.

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.