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SANTA CRUZ BIOTECHNOLOGY, INC.

VEGF siRNA (h2): sc-44278

BACKGROUND

The onset of angiogenesis is believed to be an early event in tumorigenesis and may facilitate tumor progression and metastasis. Several growth factors with angiogenic activity have been described. These include fibroblast growth factors (FGFs), platelet derived growth factor (PDGF) and vascular endothelial growth factor (VEGF). VEGF is a dimeric glycoprotein with structural homology to PDGF. Several variants of VEGF have been described that arise by alternative mRNA splicing. It has been speculated that VEGF may function as a tumor angiogenesis factor *in vivo* because the expression pattern of VEGF is consistent with a role in embryonic angiogenesis. VEGF mRNA is formed in some primary tumors, VEGF is produced by tumor cell lines *in vitro* and VEGF mitogenic activity appears to be restricted to endothelial cells. A member of the PDGF receptor family, Flt, has been identified as a high-affinity receptor for VEGF.

REFERENCES

1. Folkman, J., et al. 1989. Induction of angiogenesis during the transition from hyperplasia to neoplasia. Nature 339: 58-61.
2. Conn, G., et al. 1990. Purification of a glycoprotein vascular endothelial cell mitogen from a rat glioma-derived cell line. Proc. Natl. Acad. Sci. USA 87: 1323-1327.
3. Ferrara, N., et al. 1991. The vascular endothelial growth factor family of polypeptides. J. Cell. Biochem. 47: 211-218.
4. Tischer, E., et al. 1991. The human gene for vascular endothelial growth factor. Multiple protein forms are encoded through alternative exon splicing. J. Biol. Chem. 266: 11947-11954.
5. Plate, K.H., et al. 1992. Vascular endothelial growth factor is a potential tumour angiogenesis factor in human gliomas *in vivo*. Nature 359: 845-848.

CHROMOSOMAL LOCATION

Genetic locus: VEGFA (human) mapping to 6p21.1.

PRODUCT

VEGF 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see VEGF shRNA Plasmid (h2): sc-44278-SH and VEGF shRNA (h2) Lentiviral Particles: sc-44278-V as alternate gene silencing products.

For independent verification of VEGF (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-44278A, sc-44278B and sc-44278C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

VEGF siRNA (h2) is recommended for the inhibition of VEGF 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 µM in 66 µl. 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

VEGF (C-1): sc-7269 is recommended as a control antibody for monitoring of VEGF 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 VEGF gene expression knockdown using RT-PCR Primer: VEGF (h2)-PR: sc-44278-PR (20 µl, 516 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.