SANTA CRUZ BIOTECHNOLOGY, INC.

Vitronectin siRNA (m): sc-36821

BACKGROUND

Fibronectin and Vitronectin are extracellular matrix glycoproteins that are present on most cell surfaces, in extracellular fluids, and in plasma. Both Fibronectin and Vitronectin have been shown to be involved in various functions including cell adhesion, cell motility and wound healing. Vitronectin contains an RGD (Arg-Gly-Asp acid) sequence that is present in many cell adhesion ligands. The RGD sequence has been shown to be essential for cell adhesion. Increased expression of Vitronectin, integrins and plasminogen activators has been observed in migrating cells during wound healing. Vitronectin has been shown to enhance smooth cell migration, and PAI-1 has been shown to bind to Vitronectin with high affinity, resulting in the blocking of smooth cell migration. Glycosaminoglycans, proteins involved in the anchoring of Vitronectin to the extracellular matrix, have been shown to stimulate the cleavage of Vitronectin by plasmin. This cleavage reduces the affinity of Vitronectin for PAI-1.

REFERENCES

1. Akiyama, S.K., et al. 1981. The structure of fibronectin and its role in cellular adhesion. J. Supramol. Struct. Cell Biochem. 16: 345-348.
2. Ruoslahti, E., et al. 1982. Molecular and biological interactions in Fibronectin. J. Invest. Dermatol. 79: 65-68.
3. Chain, D., et al. 1991. Plasmin cleavage of vitronectin. Identification of the site and consequenct attenuation in binding plasminogen activator inhibitor-1. FEBS Lett. 285: 251-256.
4. Bauer, J.S., et al. 1992. Motility of Fibronectin receptor-deficient cells on Fibronectin and Vitronectin: collaborative interactions among integrins. J. Cell Biol. 116: 477-487.
5. Cherny, R.C., et al. 1993. Site-directed mutagenesis of the arginine-glycine-aspartic acid in Vitronectin abolishes cell adhesion. J. Biol. Chem. 268: 9725-9729.
6. Stefansson, S. and Lawrence, D.A. 1996. The serpin PAI-1 inhibits cell migration by blocking Integrin $\alpha_V\beta_3$ binding to Vitronectin. Nature 383: 441-443.

CHROMOSOMAL LOCATION

Genetic locus: Vtn (mouse) mapping to 11 B5.

PRODUCT

Vitronectin 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Vitronectin shRNA Plasmid (m): sc-36821-SH and Vitronectin shRNA (m) Lentiviral Particles: sc-36821-V as alternate gene silencing products.

For independent verification of Vitronectin (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-36821A, sc-36821B and sc-36821C.

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

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

Vitronectin 65/75 (D-8): sc-74484 is recommended as a control antibody for monitoring of Vitronectin 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 Vitronectin gene expression knockdown using RT-PCR Primer: Vitronectin (m)-PR: sc-36821-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

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.