



Nectin 1 siRNA (h): sc-36022

BACKGROUND

Nectin is a Ca^{2+} -independent homophilic cell adhesion molecule that belongs to the immunoglobulin superfamily. Human Nectin is identical to the poliovirus receptor-related protein (PRR) and is identified to be the α herpesvirus entry mediator. Nectin constitutes a family consisting of at least Nectin 1, 2 and 3. Nectin 2 and 3 are ubiquitously expressed, whereas Nectin 1 is abundantly expressed in the brain. Nectin 1 exists as Nectin 1 α and 1 β /HlgR, produced by alternative splicing. The cytoplasmic regions of Nectin 1 α , but not Nectin 1 β /HlgR, have a C-terminal conserved motif (E/A-X-Y-V). This motif interacts with the PDZ domain of the F-Actin-binding protein, afadin, through which it is linked to the Actin cytoskeleton. Nectin 1, also designated HveC/ PRR1, allows the entry of herpes simplex virus type 1 (HSV-1) and HSV-2 into mammalian cells. The interaction of virus envelope glycoprotein D (gD) with Nectin 1 is an essential step in the process leading to membrane fusion; the gD binding site is located at the first Ig-like domain of Nectin 1. Both the transinteraction of Nectin and the interaction of Nectin with afadin are necessary for their co-localization with E-cadherin and catenins at adherens junctions.

REFERENCES

1. Lopez, M., et al. 1995. Complementary DNA characterization and chromosomal localization of a human gene related to the poliovirus receptor-encoding gene. *Gene* 155: 261-265.
2. Geraghty, R.J., et al. 1998. Entry of α herpesviruses mediated by poliovirus receptor-related protein 1 and poliovirus receptor. *Science* 280: 1618-1620.
3. Kikyo, M., et al. 2000. Cell-cell adhesion-mediated tyrosine phosphorylation of Nectin-2 δ , an immunoglobulin-like cell adhesion molecule at adherens junctions. *Oncogene* 19: 4022-4028.
4. Krummenacher, C., et al. 2000. Localization of a binding site for herpes simplex virus glycoprotein D on herpesvirus entry mediator C by using antireceptor monoclonal antibodies. *J. Virol.* 74: 10863-10872.
5. Shukla, D., et al. 2000. Striking similarity of murine Nectin-1 α (HveC) in sequence and activity as a glycoprotein D receptor for α herpesvirus entry. *J. Virol.* 74: 11773-11781.

CHROMOSOMAL LOCATION

Genetic locus: PVRL1 (human) mapping to 11q23.3.

PRODUCT

Nectin 1 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Nectin 1 shRNA Plasmid (h): sc-36022-SH and Nectin 1 shRNA (h) Lentiviral Particles: sc-36022-V as alternate gene silencing products.

For independent verification of Nectin 1 (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-36022A, sc-36022B and sc-36022C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

Nectin 1 siRNA (h) is recommended for the inhibition of Nectin 1 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

Nectin 1 (CK6): sc-21722 is recommended as a control antibody for monitoring of Nectin 1 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 MarkerTM 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 Nectin 1 gene expression knockdown using RT-PCR Primer: Nectin 1 (h)-PR: sc-36022-PR (20 μl , 596 bp). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.