

VASP siRNA (m): sc-36809

BACKGROUND

The Wiskott-Aldrich syndrome (WAS) is characterized by thrombocytopenia, eczema, defects in cell-mediated and humoral immunity, and a propensity for lymphoproliferative diseases. The syndrome is the result of a mutation in the gene encoding a proline-rich protein termed WASP. WASP has been identified as a downstream effector of Cdc42 and has been implicated in Actin polymerization and cytoskeletal organization. A distantly related protein, VASP (vaso-dilator-stimulated phosphoprotein), is involved in the maintenance of cytoarchitecture by interacting with Actin-like filaments. VASP shares a limited degree of homology with the amino-terminus of WASP, which is frequently mutated in WAS patients. An established substrate of cAMP and cGMP dependent kinases, VASP is phosphorylated on a regulatory Serine residue 157 and localizes to focal adhesions, microfilaments and highly active regions of the plasma membrane. VASP is highly expressed in human platelets and, like WASP, may play a role in cytoskeletal organization.

REFERENCES

1. Reinhard, M., et al. 1992. The 46/50 kDa phosphoprotein VASP purified from human platelets is a novel protein associated with Actin filaments and focal contacts. *EMBO J.* 11: 2063-2070.
2. Reinhard, M., et al. 1995. Identification, purification and characterization of a Zyxin-related protein that binds the focal adhesion and microfilament protein VASP (vasodilator-stimulated phosphoprotein). *Proc. Natl. Acad. Sci. USA* 92: 7956-7960
3. Symons, M., et al. 1996. Wiskott-Aldrich syndrome protein, a novel effector for the GTPase Cdc42Hs, is implicated in actin polymerization. *Cell* 84: 723-734.

CHROMOSOMAL LOCATION

Genetic locus: Vasp (mouse) mapping to 7 A3.

PRODUCT

VASP 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 VASP shRNA Plasmid (m): sc-36809-SH and VASP shRNA (m) Lentiviral Particles: sc-36809-V as alternate gene silencing products.

For independent verification of VASP (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-36809A, sc-36809B and sc-36809C.

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

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

VASP (A-11): sc-46668 is recommended as a control antibody for monitoring of VASP 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 VASP gene expression knockdown using RT-PCR Primer: VASP (m)-PR: sc-36809-PR (20 μ l, 531 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Knauer, O., et al. 2008. Differential phosphoproteome profiling reveals a functional role for VASP in *Helicobacter pylori*-induced cytoskeleton turnover in gastric epithelial cells. *Cell. Microbiol.* 10: 2285-2296.
2. Profirovic, J., et al. 2011. Vasodilator-stimulated phosphoprotein deficiency potentiates PAR-1-induced increase in endothelial permeability in mouse lungs. *J. Cell. Physiol.* 126: 1255-1264.

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.