

WAVE3 siRNA (h): sc-44192

BACKGROUND

WASP (for Wiskott-Aldrich syndrome protein) and N-WASP are downstream effectors of Cdc42 that are implicated in Actin polymerization and cytoskeletal organization. The WASP family also includes VASP (vasodilator-stimulated phosphoprotein) and Mena (for mammalian enabled protein), which accumulate at focal adhesions and are also involved in the regulation of the Actin cytoskeleton. The WAVE proteins are related to the WASP family proteins and are likewise involved in mediating Actin reorganization downstream of the Rho family of small GTPases. The two protein homologs WAVE1 and WAVE2 specifically regulate membrane ruffling by inducing the formation of Actin filament clusters in response to GTP binding and activating Rac. The WAVE proteins mediate this Actin polymerization by cooperating with the Arp2/3 complex, a nucleation core, and thereby promoting the formation of Actin filaments. WAVE1, which is also designated SCAR (for suppressor of cAR), is expressed primarily in the brain, while WAVE2 is widely expressed with the expression highest in peripheral blood leukocytes.

REFERENCES

1. 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.
2. Miki, H., et al. 1998. WAVE, a novel WASP-family protein involved in Actin reorganization induced by Rac. *EMBO J.* 17: 6932-6941.
3. Machesky, L.M. and Insall, R.H. 1998. Scar1 and the related Wiskott-Aldrich syndrome protein, WASP, regulate the Actin cytoskeleton through the Arp2/3 complex. *Curr. Biol.* 8: 1347-1356.
4. Bear, J.E., et al. 1998. SCAR, a WASP-related protein, isolated as a suppressor of receptor defects in late *Dictyostelium* development. *J. Cell Biol.* 142: 1325-1335.
5. Rohatgi, R., et al. 1999. The interaction between N-WASP and the Arp2/3 complex links Cdc42-dependent signals to Actin assembly. *Cell* 97: 221-231.
6. Prehoda, K.E., et al. 1999. Structure of the enabled/VASP homology 1 domain-peptide complex: a key component in the spatial control of Actin assembly. *Cell* 97: 471-480.

CHROMOSOMAL LOCATION

Genetic locus: WASF3 (human) mapping to 13q12.13.

PRODUCT

WAVE3 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see WAVE3 shRNA Plasmid (h): sc-44192-SH and WAVE3 shRNA (h) Lentiviral Particles: sc-44192-V as alternate gene silencing products.

For independent verification of WAVE3 (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-44192A, sc-44192B and sc-44192C.

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

WAVE3 shRNA Plasmid (h) is recommended for the inhibition of WAVE3 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 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

WAVE3 (E-3): sc-515303 is recommended as a control antibody for monitoring of WAVE3 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 WAVE3 gene expression knockdown using RT-PCR Primer: WAVE3 (h)-PR: sc-44192-PR (20 μ l). 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.