SANTA CRUZ BIOTECHNOLOGY, INC.

SAV1 siRNA (h): sc-92380

BACKGROUND

SAV1 (Salvador homolog 1), also known as SAV, WW45 (45 kDa WW domain protein) or WWP4, is a ubiquitously expressed protein with one SARAH (Salvador/Rassf/Hippo) domain and two WW domains. The SARAH domain is a protein-protein interaction domain that is involved in cell cycle regulation and apoptosis. SAV1 can form homodimers and is believed to function as a scaffold protein of the Hippo pathway. Via its SARAH domain, SAV1 is capable of binding Krs-2, a protein that restricts cell proliferation and promotes apoptosis. This interaction is important for the transduction of apoptosis and cell cycle arrest signals. More specifically, SAV1 is essential for the nuclear translocation and activation of Krs-2. Both of these events (Krs-2 translocation and activation) are required for the subsequent phosphorylation of LATS1 and kpm, two major tumor suppressors. Defects in this pathway (the Hippo pathway) have been associated with tumorigenesis, suggesting that dysfunctional SAV1 may contribute to tumor development.

REFERENCES

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2. Tapon, N., et al. 2002. Salvador promotes both cell cycle exit and apoptosis in *Drosophila* and is mutated in human cancer cell lines. Cell 110: 467-478.
3. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 607203. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Dong, J., et al. 2007. Elucidation of a universal size-control mechanism in *Drosophila* and mammals. Cell 130: 1120-1133.
5. Guo, C., et al. 2007. RASSF1A is part of a complex similar to the *Drosophila* Hippo/Salvador/LATS tumor-suppressor network. Curr. Biol. 17: 700-705.
6. Seidel, C., et al. 2007. Frequent hypermethylation of MST1 and MST2 in soft tissue sarcoma. Mol. Carcinog. 46: 865-871.
7. Hwang, E., et al. 2007. Structural insight into dimeric interaction of the SARAH domains from MST1 and RASSF family proteins in the apoptosis pathway. Proc. Natl. Acad. Sci. USA 104: 9236-9241.

CHROMOSOMAL LOCATION

Genetic locus: SAV1 (human) mapping to 14q22.1.

PRODUCT

SAV1 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 SAV1 shRNA Plasmid (h): sc-92380-SH and SAV1 shRNA (h) Lentiviral Particles: sc-92380-V as alternate gene silencing products.

For independent verification of SAV1 (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-92380A, sc-92380B and sc-92380C.

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

SAV1 siRNA (h) is recommended for the inhibition of SAV1 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

SAV1 (F-5): sc-374366 is recommended as a control antibody for monitoring of SAV1 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 SAV1 gene expression knockdown using RT-PCR Primer: SAV1 (h)-PR: sc-92380-PR (20 µl, 573 bp). 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.