SANTA CRUZ BIOTECHNOLOGY, INC.

SAP-1 siRNA (h): sc-77346

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The Power to Question

BACKGROUND

Serum response factor (SRF) is a transcription factor that binds the serum response element (SRE), a sequence that mediates the transient response of many cellular genes to growth stimulation. SRF-binding sites are also constitutive promoter elements in many muscle-specific promoters. At the c-Fos SRE, formation of a ternary complex containing SRF and its accessory protein p62TCF appears to be important for signal transduction. Two related Ets domain proteins, Elk-1 and SRF accessory protein-1 (SAP-1), have DNA binding properties identical to that of p62TCF. Elk-1 and SAP-1 contain two homologous regions of which the two amino terminal regions, the Ets domain (box A) and the B box, mediate ternary complex formation with SRF. The third homologous region, the C box located toward the C-terminus of the proteins, contains conserved consensus phosphorylation sites for MAP kinases.

REFERENCES

1. Norman, C., et al. 1988. Isolation and properties of cDNA clones encoding SRF, a transcription factor that binds to the c-Fos serum response element. Cell 55: 989-1003.
2. Boxer, L.M., et al. 1989. The sarcomeric actin CArG-binding factor is indistinguishable from the c-Fos serum response factor. Mol. Cell. Biol. 9: 515-522.
3. Treisman, R. 1990. The SRE: a growth factor responsive transcriptional regulator. Semin. Cancer Biol. 1: 47-58.
4. Malik, R.K., et al. 1991. Epidermal growth factor and other motigens induce binding of a protein complex to the c-Fos serum response element in human astrocytoma and other cells. J. Biol. Chem. 266: 8576-8582.
5. Dalton, S., et al. 1992. Characterization of SAP-1, a protein recruited by serum response factor to the c-Fos serum response element. Cell 68: 597-612.
6. Treisman, R.H., et al. 1992. Spatial flexibility in ternary complex formation by SRF and its accessory proteins. EMBO J. 11: 4631-4640.
7. Rao, V.N., et al. 1992. Elk-1 domains responsible for autonomous DNA binding, SRE:SRF interaction and negative regulation of DNA binding. Oncogene 7: 2335-2440.
8. Hill, C.S., et al. 1993. Functional analysis of a growth factor-responsive transcription factor complex. Cell 73: 395-406.

CHROMOSOMAL LOCATION

Genetic locus: ELK4 (human) mapping to 1q32.1.

PRODUCT

SAP-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 SAP-1 shRNA Plasmid (h): sc-77346-SH and SAP-1 shRNA (h) Lentiviral Particles: sc-77346-V as alternate gene silencing products.

For independent verification of SAP-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-77346A, sc-77346B and sc-77346C.

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

SAP-1 siRNA (h) is recommended for the inhibition of SAP-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

SAP-1a (H-3): sc-166823 is recommended as a control antibody for monitoring of SAP-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-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 SAP-1 gene expression knockdown using RT-PCR Primer: SAP-1 (h)-PR: sc-77346-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.