

STAP-2 siRNA (m): sc-153877

BACKGROUND

Protein kinases comprise a large group of encoded factors that regulate cellular processes by catalyzing the transfer of a phosphate group to a hydroxyl acceptor in serine, threonine or tyrosine residues. Kinases are capable of influencing the oncogenic potential of cell systems at the level of oncoprotein or tumor suppressor protein phosphorylation states. STAP-2 is a protein that contains a pleckstrin homology (PH) domain and an SH2 domain, and associates with BRK. BRK (breast tumor kinase, Sik) is a 451 amino acid, nonreceptor protein-tyrosine kinase that is overexpressed in breast tumors and metastatic melanoma cell lines. Similar to the Src family of intracellular kinases, BRK is comprised of an SH3 domain, an SH2 domain, and a catalytic domain. STAP-2 is susceptible to tyrosine phosphorylation and may be involved in tyrosine kinase-mediated signaling cascades, whose aberrant function may lead to metastasis.

REFERENCES

1. Hunter, T. 1995. Protein kinases and phosphatases: the yin and yang of protein phosphorylation and signaling. *Cell* 80: 225-236.
2. Vasioukhin, V., et al. 1997. A role for the epithelial-cell-specific tyrosine kinase Sik during keratinocyte differentiation. *Proc. Natl. Acad. Sci. USA* 94:14477-14482.
3. Mitchell, P.J., et al. 1997. Characterisation and chromosome mapping of the human non receptor tyrosine kinase gene, brk. *Oncogene* 15: 1497-1502.
4. Mitchell, P.J., et al. 2000. A novel adaptor-like protein which is a substrate for the non-receptor tyrosine kinase, BRK. *Oncogene* 19: 4273-4282.

CHROMOSOMAL LOCATION

Genetic locus: Stap2 (mouse) mapping to 17 D.

PRODUCT

STAP-2 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 STAP-2 shRNA Plasmid (m): sc-153877-SH and STAP-2 shRNA (m) Lentiviral Particles: sc-153877-V as alternate gene silencing products.

For independent verification of STAP-2 (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-153877A, sc-153877B and sc-153877C.

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

STAP-2 siRNA (m) is recommended for the inhibition of STAP-2 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

STAP-2 (H-10): sc-514716 is recommended as a control antibody for monitoring of STAP-2 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor STAP-2 gene expression knockdown using RT-PCR Primer: STAP-2 (m)-PR: sc-153877-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 or our catalog for detailed protocols and support products.