

SCYL2 siRNA (m): sc-153279

BACKGROUND

SCYL2 (SCY1-like 2), also known as CVAK104 (Coated vesicle-associated kinase of 104 kDa), is a 929 amino acid protein that contains one HEAT repeat and one protein kinase domain. Localized to a variety of locations within the cell, including the perinuclear region of the cytoplasm, the *trans*-golgi network membrane and clathrin-coated vesicles, SCYL2 exists as a component of AP-2-containing Clathrin coated structures. SCYL2 is thought to function as a Ser/Thr protein kinase, exhibiting kinase activity towards adaptor complex proteins, possibly regulating Clathrin-dependent trafficking events between the Golgi network and endosomes. Additionally, SCYL2 may play a role in the sorting of SNARE proteins and, ultimately, in embryonic development.

REFERENCES

1. Hanks, S.K., et al. 1988. The protein kinase family: conserved features and deduced phylogeny of the catalytic domains. *Science* 241: 42-52.
2. Nagase, T., et al. 2000. Prediction of the coding sequences of unidentified human genes. XVI. The complete sequences of 150 new cDNA clones from brain which code for large proteins *in vitro*. *DNA Res.* 7: 65-73.
3. Conner, S.D., et al. 2005. CVAK104 is a novel poly-L-lysine-stimulated kinase that targets the β 2-subunit of AP-2. *J. Biol. Chem.* 280: 21539-21544.
4. Düwel, M., et al. 2006. Clathrin-dependent association of CVAK104 with endosomes and the *trans*-Golgi network. *Mol. Biol. Cell* 17: 4513-4525.
5. Schmid, E.M., et al. 2006. Role of the AP-2 β -appendage hub in recruiting partners for Clathrin-coated vesicle assembly. *PLoS Biol.* 4: e262.
6. Borner, G.H., et al. 2007. CVAK104 is a novel regulator of Clathrin-mediated SNARE sorting. *Traffic* 8: 893-903.

CHROMOSOMAL LOCATION

Genetic locus: Scyl2 (mouse) mapping to 10 C2.

PRODUCT

SCYL2 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 SCYL2 shRNA Plasmid (m): sc-153279-SH and SCYL2 shRNA (m) Lentiviral Particles: sc-153279-V as alternate gene silencing products.

For independent verification of SCYL2 (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-153279A, sc-153279B and sc-153279C.

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.

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

SCYL2 siRNA (m) is recommended for the inhibition of SCYL2 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

SCYL2 (B-3): sc-515916 is recommended as a control antibody for monitoring of SCYL2 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 SCYL2 gene expression knockdown using RT-PCR Primer: SCYL2 (m)-PR: sc-153279-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.