



Asef siRNA (m): sc-41697

BACKGROUND

The adenomatous polyposis coli gene (APC) is mutated in familial adenomatous polyposis and in sporadic colorectal tumors. Familial adenomatous polyposis is a dominantly inherited disease characterized by multiple adenomatous polyps in the colon. The human full-length Asef cDNA encodes a protein of 619 amino acids with motifs found in the Dbl family of proteins. The APC gene product binds through its armadillo repeat domain to a Rac-specific guanine nucleotide exchange factor (GEF), termed APC-stimulated guanine nucleotide exchange factor (Asef). Endogenous APC has been shown to co-localize with Asef in mouse colon epithelial cells and neuronal cells. Furthermore, APC has been shown to enhance the GEF activity of Asef and stimulate Asef-mediated cell flattening, membrane ruffling and lamellipodia formation in MDCK cells, suggesting that the APC-Asef complex may regulate the actin cytoskeletal network, cell morphology and migration, and neuronal function. Full-length APC protein is located in both the membrane/cytoskeletal and the nuclear fractions.

REFERENCES

1. Kinzler, K.W. and Vogelstein, B. 1996. Lessons from hereditary colorectal cancer. *Cell* 87: 159-170.
2. Cerione, R.A. and Zheng, Y. 1996. The Dbl family of oncogenes. *Curr. Opin. Cell Biol.* 8: 216-222.
3. Whitehead, I.P., Campbell, S., Rossman, K.L. and Der, C.J. 1997. Dbl family proteins. *Biochim. Biophys. Acta* 1332: F1-F23.
4. Neufeld, K.L. and White, R.L. 1997. Nuclear and cytoplasmic localizations of the adenomatous polyposis coli protein. *Proc. Natl. Acad. Sci. USA* 94: 3034-3039.
5. Polakis, P. 1997. The adenomatous polyposis coli (APC) tumor suppressor. *Biochim. Biophys. Acta* 1332: F127-F147.
6. Bienz, M. 1999. APC: the plot thickens. *Curr. Opin. Genet. Dev.* 9: 595-603.
7. Kawasaki, Y., Senda, T., Ishidate, T., Koyama, R., Morishita, T., Iwayama, Y., Higuchi, O. and Akiyama, T. 2000. Asef, a link between the tumor suppressor APC and G protein signaling. *Science* 289: 1194-1197.
8. Kawasaki, Y. and Akiyama, T. 2001. A novel function of the tumor suppressor APC. *Tanpakushitsu Kakusan Koso* 46: 228-232.

CHROMOSOMAL LOCATION

Genetic locus: Arhgef4 (mouse) mapping to 1 B.

PRODUCT

Asef 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 Asef shRNA Plasmid (m): sc-41697-SH and Asef shRNA (m) Lentiviral Particles: sc-41697-V as alternate gene silencing products.

For independent verification of Asef (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-41697A, sc-41697B and sc-41697C.

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

Asef siRNA (m) is recommended for the inhibition of Asef 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Asef gene expression knockdown using RT-PCR Primer: Asef (m)-PR: sc-41697-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.