SANTA CRUZ BIOTECHNOLOGY, INC.

EB3 siRNA (h): sc-37608

BACKGROUND

EB1 (MAPRE2, microtubule-associated protein, RP/EB family, member 2, EB2, RP1) may influence tumorigenesis of colorectal cancers and proliferative control of normal cells. EB1 may belong to the intermediate/early gene family, involved in the signal transduction cascade downstream of the TCR. Colorectal cancer is caused by the pathologic transformation of normal colonic epithelium to an adenomatous polyp, which can become an invasive cancer. APC (adenomatous polyposis coli) is a tumor suppressor gene, the mutation of which is one of the earliest events in colorectal carcinogenesis. A majority of the mutations result in the loss of the carboxy terminus of APC. EB1 has been shown to bind to the carboxy terminal region of APC, which implicates EB1 in APC suppression of colonic cancer. EB1 overexpression may play a role in the development of human esophageal squamous cell carcinoma (ESCC) by affecting APC function and activating the β-catenin/TCF pathway. EB3 is related to EB1 and likewise associates with the microtubule cytoskeleton. EB3 is expressed predominantly in the central nervous system and preferentially associates with APCL.

REFERENCES

1. Cottrell, S., et al. 1992. Molecular analysis of APC mutations in familial adenomatous polyposis and sporadic colon carcinomas. Lancet 340: 626-630.
2. Smith, K.J., et al. 1993. The APC gene product in normal and tumor cells. Proc. Natl. Acad. Sci. USA 90: 2846-2850.
3. Levy, D.B., et al. 1994. Inactivation of both APC alleles in human and mouse tumors. Cancer Res. 54: 5953-5958.
4. Su, L.K., et al. 1995. APC binds to the novel protein EB1. Cancer Res. 55: 2972-2977.
5. Oda, H., et al. 1996. Somatic mutations of the APC gene in sporadic hepatoblastomas. Cancer Res. 56: 3320-3323.
6. Gryfe, R., et al. 1997. Molecular biology of colorectal cancer. Curr. Probl. Cancer 21: 233-300.

CHROMOSOMAL LOCATION

Genetic locus: MAPRE3 (human) mapping to 2p23.3.

PRODUCT

EB3 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 EB3 shRNA Plasmid (h): sc-37608-SH and EB3 shRNA (h) Lentiviral Particles: sc-37608-V as alternate gene silencing products.

For independent verification of EB3 (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-37608A, sc-37608B and sc-37608C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

EB3 siRNA (h) is recommended for the inhibition of EB3 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

EB3 (7): sc-136405 is recommended as a control antibody for monitoring of EB3 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 EB3 gene expression knockdown using RT-PCR Primer: EB3 (h)-PR: sc-37608-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.