SANTA CRUZ BIOTECHNOLOGY, INC.

gankyrin siRNA (h): sc-72186

BACKGROUND

Gankyrin, a hepatocellular carcinoma-associated protein, regulates protein-protein interactions in cell cycle control as well as protein degradation. Furthermore, upregulation of gankyrin correlates with cell-cycle progression in normal hepatocytes as well. It contains six domains known as ankyrin repeats, and interacts with Rb, Cdk4, the 26S proteasome, and MAGE-A4. This last interaction suppresses anchorage-independent growth in gankyrin overexpressing cells, demonstrating a possible mechanism for immunotherapy in hepatocellular carcinoma.

REFERENCES

1. Iwai, A., et al. 2003. Role of a novel oncogenic protein, gankyrin, in hepatocyte proliferation. J. Gastroenterol. 38: 751-758.
2. Nagao, T., et al. 2003. MAGE-A4 interacts with the liver oncoprotein gankyrin and suppresses its tumorigenic activity. J. Biol. Chem. 278: 10668-10674.
3. Krzywda, S., et al. 2004. The crystal structure of gankyrin, an oncoprotein found in complexes with cyclin-dependent kinase 4, a 19S proteasomal ATPase regulator, and the tumor suppressors Rb and p53. J. Biol. Chem. 279: 1541-1545.
4. Higashitsuji, H., et al. 2005. The oncoprotein gankyrin binds to MDM2/HDM2, enhancing ubiquitylation and degradation of p53. Cancer Cell 8: 75-87.
5. Higashitsuji, H., et al. 2007. The oncoprotein gankyrin interacts with RelA and suppresses NFκB activity. Biochem. Biophys. Res. Commun. 363: 879-884.

CHROMOSOMAL LOCATION

Genetic locus: PSMD10 (human) mapping to Xq22.3.

PRODUCT

gankyrin 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 gankyrin shRNA Plasmid (h): sc-72186-SH and gankyrin shRNA (h) Lentiviral Particles: sc-72186-V as alternate gene silencing products.

For independent verification of gankyrin (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-72186A, sc-72186B and sc-72186C.

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

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

gankyrin (3A6C2): sc-101498 is recommended as a control antibody for monitoring of gankyrin 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 gankyrin gene expression knockdown using RT-PCR Primer: gankyrin (h)-PR: sc-72186-PR (20 µl, 530 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Li, J., et al. 2011. Gankyrin, a biomarker for epithelial carcinogenesis, is overexpressed in human oral cancer. Anticancer Res. 31: 2683-2692.
2. Sakurai, T., et al. 2017. Gankyrin induces STAT3 activation in tumor microenvironment and sorafenib resistance in hepatocellular carcinoma. Cancer Sci. 108: 1996-2003.

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.