SANTA CRUZ BIOTECHNOLOGY, INC.

p15 INK4B siRNA (h): sc-37624

BACKGROUND

The normal progression of cells through the cell cycle is under the control of the cyclin dependent protein kinases Cdk4 and Cdk6, which are subject to inhibition by the mitotic inhibitory protein, p16 INK4A. An isolated member of the p16 INK4A family has been designated p15 INK4B (also designated, p15, INK4B, CDK4I, TP15, or MTS2). p15 INK4B expression is upregulated approximately 30-fold in TGFβ-treated human keratinocytes, suggesting that p15 INK4B may act as an effector of TGFβ-mediated cell cycle arrest. The gene encoding p15 INK4B (CDKN2B) has been mapped to chromosome 9p21.3 at a position adjacent to the p16 INK4A gene at a site of frequent chromosomal abnormality in human tumors. It has been suggested that p15 INK4B may function as an effector of TGFβ-mediated cell cycle arrest through inhibition of Cdk4 and Cdk6 kinases.

REFERENCES

1. Hannon, G.J., et al. 1994. p15 INK4B is a potential effector of TGFβ-induced cell cycle arrest. Nature 371: 257-261.
2. Kamb, A., et al. 1994. A cell cycle regulator potentially involved in genesis of many tumor types. Science 264: 436-440.
3. Reynisdóttir, I., et al. 1997. The subcellular locations of p15 INK4B and $p27^{Kip1}$ coordinate their inhibitory interactions with Cdk4 and Cdk2. Genes Dev. 11: 492-503.
4. Kiyota, A., et al. 2002. Anti-epidermal growth factor receptor monoclonal and p15 INK4B and induces G_1 arrest in oral squamous carcinoma cell lines. Oncology 63: 92-98.

CHROMOSOMAL LOCATION

Genetic locus: CDKN2B (human) mapping to 9p21.3.

PRODUCT

p15 INK4B 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 p15 INK4B shRNA Plasmid (h): sc-37624-SH and p15 INK4B shRNA (h) Lentiviral Particles: sc-37624-V as alternate gene silencing products.

For independent verification of p15 INK4B (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-37624A, sc-37624B and sc-37624C.

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

p15 INK4B siRNA (h) is recommended for the inhibition of p15 INK4B 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

p15 INK4B (D-12): sc-271791 is recommended as a control antibody for monitoring of p15 INK4B 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 p15 INK4B gene expression knockdown using RT-PCR Primer: p15 INK4B (h)-PR: sc-37624-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Choi, S., et al. 2009. Ginsenoside Rh2-mediated G_1 phase cell cycle arrest in human breast cancer cells is caused by p15 INK4B and $p27^{Kip1}$-dependent inhibition of cyclin-dependent kinases. Pharm. Res. 26: 2280-2288.
2. Lee, H.A., et al. 2021. Histone deacetylase inhibitor-induced CDKN2B and CDKN2D contribute to G_2/M cell cycle arrest incurred by oxidative stress in Hepatocellular carcinoma cells via forkhead box M1 suppression. J. Cancer 12: 5086-5098.

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.