SANTA CRUZ BIOTECHNOLOGY, INC.

cyclin D2 siRNA (h): sc-35134

BACKGROUND

The proliferation of eukaryotic cells is controlled at specific points in the cell cycle, particularly at the G_1 to S and the G_2 to M transitions. It is well established that the Cdc2 p34-cyclin B protein kinase plays a critical role in the G_2 to M transition while cyclin A associates with Cdk2 p33 and functions in S phase. Considerable effort directed towards the identification of G_1 cyclins has led to the isolation of cyclin D, cyclin C and cyclin E. Of these, cyclin D corresponds to a putative human oncogene, designated PRAD1, which maps at the site of the Bcl-1 rearrangement in certain lymphomas and leukemias. Two additional human type D cyclins, as well as their mouse homologs, have been identified. Evidence has established that members of the cyclin D family function to regulate phosphorylation of the retinoblastoma gene product, thereby activating E2F transcription factors.

REFERENCES

1. Draetta, G. 1990. Cell cycle control in eukaryotes: molecular mechanisms of cdc2 activation. Trends Biol. Sci. 15: 378-383.
2. Xiong, Y., et al. 1992. Molecular cloning and chromosomal mapping of CCND genes encoding human D-type cyclins. Genomics 13: 575-584.
3. Kiyokawa, H., et al. 1992. Cloning of a D-type cyclin from murine erythroleukemia cells. Proc. Natl. Acad. Sci. USA 89: 2444-2447.
4. Won, K., et al. 1992. Growth-regulated expression of D-type cyclin genes in human diploid fibroblasts. Proc. Natl. Acad. Sci. USA 89: 9910-9914.
5. Motokura, T., et al. 1992. Cloning and characterization of human cyclin D3, a cDNA closely related in sequence to the PRAD1/cyclin D1 proto-oncogene. J. Biol. Chem. 267: 20412-20415.

CHROMOSOMAL LOCATION

Genetic locus: CCND2 (human) mapping to 12p13.32.

PRODUCT

cyclin D2 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 cyclin D2 shRNA Plasmid (h): sc-35134-SH and cyclin D2 shRNA (h) Lentiviral Particles: sc-35134-V as alternate gene silencing products.

For independent verification of cyclin D2 (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-35134A, sc-35134B and sc-35134C.

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

Cyclin D2 shRNA Plasmid (h) is recommended for the inhibition of cyclin D2 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

cyclin D2 (B-6): sc-376676 is recommended as a control antibody for monitoring of cyclin D2 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 cyclin D2 gene expression knockdown using RT-PCR Primer: cyclin D2 (h)-PR: sc-35134-PR (20 µl, 406 bp). 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.