<u>6477</u>

SignalSilence® Cyclin D1 siRNA II (Mouse Specific)

 10 μM in 300 μl (100 transfections)



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rev. 02/11/16

For Research Use Only. Not For Use In Diagnostic Procedures.

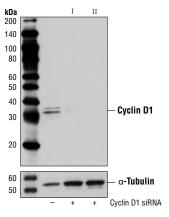
Species Cross-Reactivity: M

Description: SignalSilence[®] Cyclin D1 siRNA II (Mouse Specific) from Cell Signaling Technology (CST) allows the researcher to specifically inhibit Cyclin D1 expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules into the cell. All SignalSilence[®] siRNA products from CST are rigorously tested in-house and have been shown to reduce target protein expression by western analysis.

Background: Activity of the cyclin-dependent kinases CDK4 and CDK6 is regulated by T-loop phosphorylation, by the abundance of their cyclin partners (the D-type cyclins), and by association with CDK inhibitors of the Cip/Kip or INK family of proteins (1). The inactive ternary complex of cyclin D/CDK4 and p27 Kip1 requires extracellular mitogenic stimuli for the release and degradation of p27 concomitant with a rise in cyclin D levels to effect progression through the restriction point and pRb-dependent entry into S-phase (2). The active complex of cyclin D/CDK4 targets the retinoblastoma protein for phosphorylation, allowing the release of E2F transcription factors that activate G1/S-phase gene expression (3). Levels of cyclin D protein drop upon withdrawal of growth factors through downregulation of its protein expression and through phosphorylation-dependent degradation (4).

Directions for Use: CST recommends transfection with 100 nM SignalSilence[®] Cyclin D1 siRNA II (Mouse Specific) 48 to 72 hours prior to cell lysis. For transfection procedure, follow protocol provided by the transfection reagent manufacturer. Please feel free to contact CST with any questions on use.

Quality Control: Oligonucleotide synthesis is monitored base by base through trityl analysis to ensure appropriate coupling efficiency. The oligo is subsequently purified by affinity-solid phase extraction. The annealed RNA duplex is further analyzed by mass spectrometry to verify the exact composition of the duplex. Each lot is compared to the previous lot by mass spectrometry to ensure maximum lot-to-lot consistency.



Western blot analysis of extracts from C2C12 cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® Cyclin D1 siRNA I (Mouse Specific) (+), or SignalSilence® Cyclin D1 siRNA II (Mouse Specific) #6477 (+), using Cyclin D1 (92G2) Rabbit mAb #2978 (upper) or α -Tubulin (11H10) Rabbit mAb #2125 (lower). The Cyclin D1 (92G2) Rabbit mAb confirms silencing of Cyclin D1 expression, while the α -Tubulin (11H10) Rabbit mAb is used as a loading control.

Entrez-Gene ID #12443 Swiss-Prot Acc. #P25322

Storage: Cyclin D1 siRNA II (Mouse Specific) is supplied in RNAse-free water. *Aliquot and store at -20°C.*

Please visit www.cellsignal.com for a complete listing of recommended companion products. Background References:

(1) Hirai, H. et al. (1995) Mol. Cell. Biol. 15, 2672-2681.

(2) Sherr, C.J. (1996) Science 274, 1672-1677.

(3) Lukas, J. et al. (1996) Mol. Cell. Biol. 16, 6917-6925.

(4) Diehl, J.A. et al. (1997) Genes Dev. 11, 957-972.

 Applications Key:
 W—Western
 IP—Immunoprecipitation
 IHC—Immunohistochemistry
 ChIP—Chromatin Immunoprecipitation
 IF—Immunofluorescence
 F—Flow cytometry
 E-P—ELISA-Peptide

 Species Cross-Reactivity Key:
 H—human
 M—mouse
 R—rat
 Hm—hamster
 Mk—monkey
 Mi—mink
 C—chicken
 Dm—D. melanogaster
 X—zebrafish
 B—bovine

 Dg—dog
 Pg—pig
 Sc—S. cerevisiae
 Ce-C. elegans
 Hr—Horse
 All—all species expected
 Species enclosed in parentheses are predicted to react based on 100% homology.