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New 12/13

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

Species Cross-Reactivity: H

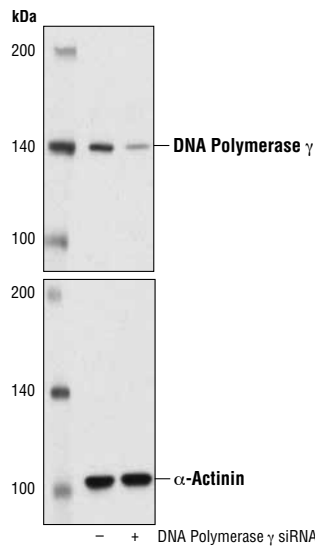
Description: SignalSilence® DNA Polymerase γ siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit DNA polymerase γ expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules 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: DNA polymerase γ (POLG, pol gamma) is a nuclear encoded protein that is responsible for mitochondrial genome replication in eukaryotic cells. The 140 kDa polymerase γ catalytic subunit forms a holoenzyme complex with a 55 kDa accessory protein (POLG2, pol γ B) dimer, which confers processivity (1). In addition to polymerase activity, polymerase γ contains 3'-5' exonuclease activity for proofreading and 5'-deoxyribonucleic phosphate lyase activity that functions in DNA base excision repair (BER). The rate at which the catalytic subunit recognizes damaged DNA during DNA repair is enhanced by the pol γ B accessory subunit (2). Mutations in the corresponding *POLG* gene are associated with several inherited neuropathies including progressive external ophthalmoplegia, myocerebro-hepatopathy spectrum disorders and Alpers-Huttenlocher syndrome (3,4). Research studies indicate that mutations in the corresponding *POLG* gene may promote breast tumorigenesis (5).

Directions for Use: CST recommends transfection with 100 nM SignalSilence® DNA Polymerase γ siRNA I 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.

Each vial contains the equivalent of 100 transfections, which corresponds to a final siRNA concentration of 100 nM per transfection in a 24-well plate with a total volume of 300 μ l per well.

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 293T cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-) or SignalSilence® DNA Polymerase γ siRNA I (+), using DNA Polymerase γ (D1Y6R) Rabbit mAb #13609 (upper) or α -Actinin (D6F6) XP® Rabbit mAb #6487 (lower). The DNA Polymerase γ (D1Y6R) Rabbit mAb confirms silencing of DNA polymerase γ expression, while the α -Actinin (D6F6) XP® Rabbit mAb is used as a loading control.

Entrez-Gene ID #5428
UniProt Acc. #P54098

Storage: DNA Polymerase γ siRNA is supplied in RNase-free water. Aliquot and store at -20°C.

For product specific protocols please see the web page for this product at www.cellsignal.com.

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

Background References:

- (1) Kaguni, L.S. (2004) *Annu Rev Biochem* 73, 293-320.
- (2) Pinz, K.G. and Bogenhagen, D.F. (2006) *DNA Repair (Amst)* 5, 121-8.
- (3) Stumpf, J.D. et al. (2013) *Cold Spring Harb Perspect Biol* 5, a011395.
- (4) Finsterer, J. (2011) *J Neurol Sci* 304, 9-16.
- (5) Singh, K.K. et al. (2009) *J Hum Genet* 54, 516-24.