

#6288 Store at -20°C

# SignalSilence® ATR siRNA I



✓ 10µM in 300 µl (100 transfections)

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rev. 02/09/16

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

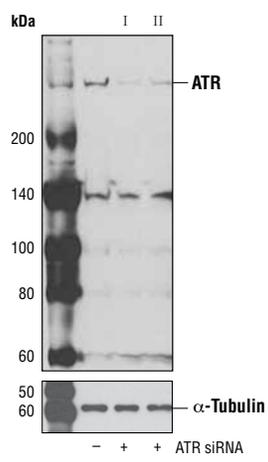
### Species Cross-Reactivity: H

**Description:** SignalSilence® ATR siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit ATR 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.

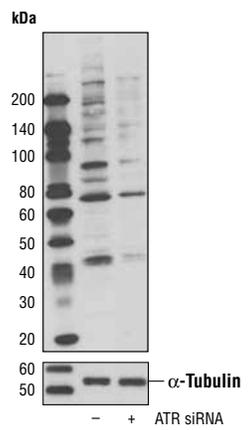
**Background:** Ataxia telangiectasia mutated kinase (ATM) and ataxia telangiectasia and Rad3-related kinase (ATR) are PI-3 Kinase-related kinase (PIKK) family members that phosphorylate multiple substrates on serine or threonine residues that are followed by a glutamine in response to DNA damage or replication blocks (1-3). Despite the essential role of ATR in cell cycle signaling and DNA repair processes, little is known about its activation. While there have been no published reports of phosphorylation sites on ATR, Cell Signaling Technology has produced an antibody directed against phospho-ATR (Ser428) that demonstrates in vivo and UV-induced phosphorylation of this protein. This reagent could prove to be a valuable tool for monitoring ATR activation. Proline-directed phosphorylation sites like this one are often targeted by CDKs and MAPKs and can often dramatically affect protein conformation (4,5).

**Directions for Use:** CST recommends transfection with 100 nM ATR 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.

**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 HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® ATR siRNA I (+) or SignalSilence® ATR siRNA II #6289 (+), using ATR Antibody #2790 (upper) or α-Tubulin (11H10) Rabbit mAb #2125 (lower). The ATR Antibody confirms silencing of ATR expression, while the α-Tubulin (11H10) Rabbit mAb is used as a loading control.



Western blot analysis of extracts from UV treated HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-) or SignalSilence® ATR siRNA I (+), using Phospho-(Ser/Thr) ATM/ATR Substrate (4F7) Rabbit mAb #2909 (upper) or α-Tubulin (11H10) Rabbit mAb #2125 (lower). The Phospho-(Ser/Thr) ATM/ATR Substrate (4F7) Rabbit mAb confirms reduction of ATR kinase activity, while the α-Tubulin (11H10) Rabbit mAb is used as a loading control.

Entrez-Gene ID #2064  
Swiss-Prot Acc. #P04626

**Storage:** ATR siRNA I is supplied in RNase-free water. Aliquot and store at -20°C.

Please visit [www.cellsignal.com](http://www.cellsignal.com) for a complete listing of recommended companion products.

### Background References:

- (1) Kastan, M.B. and Lim, D.S. (2000) *Nat. Rev. Mol. Cell Biol.* 1, 179-186.
- (2) Abraham, R.T. (2004) *DNA Repair (Amst)* 3, 883-887.
- (3) Shechter, D. et al. (2004) *DNA Repair (Amst)* 3, 901-908.
- (4) Pinna, L.A. and Ruzzene, M. (1996) *Biochim. Biophys. Acta* 1314, 191-225.
- (5) Zhou, X.Z. et al. (1999) *Cell Mol. Life Sci.* 56, 788-806.

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**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—Xenopus Z—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.