

#6302 Store at -20°C

# SignalSilence® FoxO3a siRNA I



✓ 10 µM in 300 µl (100 transfections)

Orders ■ 877-616-CELL (2355) orders@cellsignal.com  
Support ■ 877-678-TECH (8324) info@cellsignal.com  
Web ■ www.cellsignal.com

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

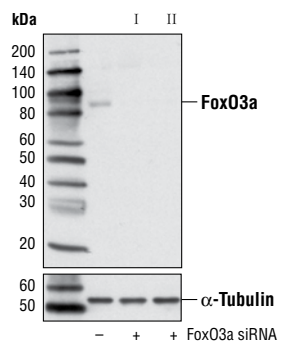
### Species Cross-Reactivity: H

**Description:** SignalSilence® FoxO3a siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit FoxO3a 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:** The Forkhead family of transcription factors is involved in tumorigenesis of rhabdomyosarcoma and acute leukemias (1-3). Within the family, three members (FoxO1, FoxO4 and FoxO3a) have sequence similarity to the nematode orthologue DAF-16, which mediates signaling via a pathway involving IGF1R, PI3K and Akt (4-6). Active forkhead members act as tumor suppressors by promoting cell cycle arrest and apoptosis. Increased expression of any FoxO member results in the activation of the cell cycle inhibitor p27Kip1. Forkhead transcription factors also play a part in TGF-β-mediated upregulation of p21CIP1, a process negatively regulated through PI3K (7). Increased proliferation results when forkhead transcription factors are inactivated through phosphorylation by Akt at Thr24, Ser256 and Ser319, which results in nuclear export and inhibition of transcription factor activity (8). Forkhead transcription factors can also be inhibited by the deacetylase sirtuin (SirT1) (9).

**Directions for Use:** CST recommends transfection with 100 nM FoxO3a 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® FoxO3a siRNA I (+) or SignalSilence® FoxO3a siRNA II #6303 (+), using FoxO3a (75D8) Rabbit mAb #2497 (upper) or α-Tubulin (11H10) Rabbit mAb #2125 (lower). The FoxO3a (75D8) Rabbit mAb confirms silencing of FoxO3a expression, while the α-Tubulin (11H10) Rabbit mAb is used as a loading control.

Entrez-Gene ID #2309  
Swiss-Prot Acc. #043524

**Storage:** FoxO3a siRNA I is supplied in RNase-free water. Aliquot and store at -20°C.

### Background References:

- (1) Anderson, M.J. et al. (1998) *Genomics* 47, 187-199.
- (2) Galili, N. et al. (1993) *Nat. Genet.* 5, 230-235.
- (3) Borkhardt, A. et al. (1997) *Oncogene* 14, 195-202.
- (4) Nakae, J. et al. (1999) *J. Biol. Chem.* 274, 15982-15985.
- (5) Rena, G. et al. (1999) *J. Biol. Chem.* 274, 17179-17183.
- (6) Guo, S. et al. (1999) *J. Biol. Chem.* 274, 17184-17192.
- (7) Seoane, J. et al. (2004) *Cell* 117, 211-223.
- (8) Arden, K.C. (2004) *Mol. Cell* 14, 416-418.
- (9) Yang, Y. et al. (2005) *EMBO J.* 24, 1021-1032.

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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.