Store at -20°C

6287

## SignalSilence® Atg14 siRNA II

300 μl
 (100 transfections)

This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

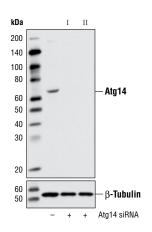
## Species Cross-Reactivity: H

**Description:** SignalSilence® Atg14 siRNA II from Cell Signaling Technology (CST) allows the researcher to specifically inhibit Atg14 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: Autophagy is a catabolic process for the autophagosomic-lysosomal degradation of bulk cytoplasmic contents (1,2). Autophagy is generally activated by conditions of nutrient deprivation but is also associated with a number of physiological processes including development, differentiation, neurodegeneration, infection and cancer (3). The molecular machinery of autophagy was largely discovered in yeast and is directed by a number of autophagy-related (Atg) genes. These proteins are involved in the formation of autophagosomes, cytoplasmic vacuoles that are delivered to lysosomes for degradation. The class III type phosphoinositide 3-kinase (PI3K) Vps34 regulates vacuolar trafficking and autophagy (4,5). Multiple proteins associate with Vsp34, including p105/Vsp15, Beclin-1, UVRAG, Atg14, and Rubicon, to determine Vsp34 function (6-12). Atg14 and Rubicon were identified based on their ability to bind to Beclin-1 and participate in unique complexes with opposing functions (9-12). Rubicon, which localizes to the endosome and lysosome, inhibits Vps34 lipid kinase activity; knockdown of Rubicon enhances autophagy and endocytic trafficking (11,12). In contrast, Atg14 localizes to autophagosomes, isolation membranes and ER, and can enhance Vps34 activity. Knockdown of Atg14 inhibits starvation-induced autophagy (11,12).

**Directions for Use:** CST recommends transfection with 100 nM Atg14 siRNA II 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.



New 08/10

Western blot analysis of extracts from HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® Atg14 siRNA I #6286 (+) or SignalSilence® Atg14 siRNA II (+), using Atg14 Antibody #5504 (upper) or  $\beta$ -Tubulin (9F3) Rabbit mAb #2128 (lower). The Atg14 Antibody confirms silencing of Atg14 expression, while the  $\beta$ -Tubulin (9F3) Rabbit mAb is used as a loading control. Entrez-Gene ID #22863 Swiss-Prot Acc. #Q6ZNS5

**Storage:** Atg14 siRNA II is supplied in RNAse-free water. Aliquot and store at -20°C.

Cell Signaling

Orders 877-616-CELL (2355)

Support 
877-678-TECH (8324)

Web www.cellsignal.com

orders@cellsignal.com

info@cellsignal.com

TECHNOLOGY®

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

## **Background References:**

- (1) Reggiori, F. and Klionsky, D.J. (2002) *Eukaryot Cell* 1, 11-21.
- (2) Codogno, P. and Meijer, A.J. (2005) *Cell Death Differ* 12 Suppl 2, 1509-18.
- (3) Levine, B. and Yuan, J. (2005) J Clin Invest 115, 2679-88.
- (4) Corvera, S. (2001) Traffic 2, 859-66.
- (5) Yan, Y. and Backer, J.M. (2007) *Biochem Soc Trans* 35, 239-41.
- (6) Stack, J.H. et al. (1995) J Cell Biol 129, 321-34.
- (7) Zeng, X. et al. (2006) J Cell Sci 119, 259-70.
- (8) Liang, C. et al. (2006) Nat Cell Biol 8, 688-99.
- (9) Itakura, E. et al. (2008) Mol Biol Cell 19, 5360-72.
- (10) Sun, Q. et al. (2008) *Proc Natl Acad Sci U S A* 105, 19211-6.
- (11) Zhong, Y. et al. (2009) Nat Cell Biol 11, 468-76.
- (12) Matsunaga, K. et al. (2009) Nat Cell Biol 11, 385-96.

 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

 Dm—don
 Pa—inin
 Species enclosed in parentheses are predicted to react based on 100% homology.
 Him—Horse
 AII—all species expected
 Species enclosed in parentheses are predicted to react based on 100% homology.