SignalSilence® Atg14 siRNA I

Storage:

Storage: Atg14 siRNA I is supplied in RNase-free water. Aliquot and store at -20°C.

Please visit www.cellsignal.com for a complete listing and store at -20°C.

Species Cross-Reactivity: H
Description: SignalSilence® Atg14 siRNA I 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 autophagosomal-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 lipid kinase activity; knockdown of Atg14 localizes to the endosome and lysosome, inhibits Vps34 lipid kinase activity; knockdown of Rubicon localizes to autophagosomes, isolation membranes and ER, and can enhance Vps34 activity. Knockdown of Atg14 inhibits starvation-induced autophagy (11,12). Rubicon, which associates 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 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.

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

Species Cross-Reactivity Key:
H—human
M—mouse
R—rat
Hm—hamster
Mm—monkey
Mg—pig
C—chicken
Dm—D. melanogaster
X—Xenopus
Z—zebrafish
B—bovine
Sn—snake
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.

Entrez-Gene ID: #22863
Swiss-Prot Acc.: #Q6XNE5

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

Background References:

Western blot analysis of extracts from HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® Atg14 siRNA I (+) or SignalSilence® Atg14 siRNA II #6287 (+), using Atg14 Antibody #5504 (upper) and β-Tubulin (9F3) Rabbit mAb is used as a loading control.