

SignalSilence® LC3A siRNA I



✓ 10 µM in 300 µl (100 transfections)

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

Species Cross-Reactivity: H, (M, R)

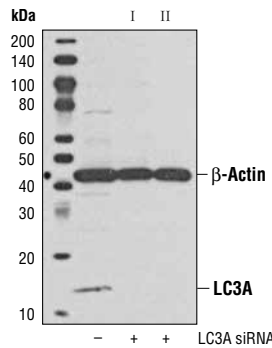
Description: SignalSilence® LC3A siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit LC3A expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules into the cell. LC3A siRNA I specifically inhibits LC3A expression and is not expected to inhibit expression of related LC3B or LC3C isoforms. 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 has also been associated with a number of physiological processes including development, differentiation, neurodegenerative diseases, infection and cancer (3). Autophagy marker Light Chain 3 (LC3) was originally identified as a subunit of microtubule-associated proteins 1A and 1B (termed MAP1LC3) (4), and subsequently found to contain similarity to the yeast protein Apg8/Aut7/Cvt5 critical for autophagy (5). Three human LC3 isoforms (LC3A, LC3B, and LC3C) undergo post-translational modifications during autophagy (6-9). Cleavage of LC3 at the carboxy terminus immediately following synthesis yields the cytosolic LC3-I form. During autophagy, LC3-I is converted to LC3-II through lipidation by a ubiquitin-like system involving Atg7 and Atg3 that allows for LC3 to become associated with autophagic vesicles (6-10). The presence of LC3 in autophagosomes and the conversion of LC3 to the lower migrating form LC3-II have been used as indicators of autophagy (11).

Directions for Use: CST recommends transfection with 100 nM LC3A 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.

Specificity/ Sensitivity: SignalSilence® LC3A siRNA I will inhibit human, mouse and rat LC3A expression.



Western blot analysis of extracts from HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® LC3A siRNA I (+) or SignalSilence® LC3A siRNA II #6215 (+), using LC3B (G40) Antibody #4108 and β-Actin (13E5) Rabbit mAb #4970. The LC3B (G40) antibody confirms silencing of LC3A expression, while the β-Actin (13E5) Rabbit mAb is used to control for loading and specificity of LC3A siRNA. Please note #4108 recognizes LC3A and LC3C.

Entrez-Gene ID #84557
Swiss-Prot Acc. #Q9H492

Storage: LC3A siRNA I is supplied in RNase-free water. Aliquot and store at -20°C.

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, 1509-1518.
- (3) Levine, B. and Yuan, J. (2005) *J. Clin. Invest.* 115, 2679-2688.
- (4) Mann, S.S. and Hammarback, J.A. (1994) *J. Biol. Chem.* 269, 11492-11497.
- (5) Lang, T. et al. (1998) *EMBO J.* 17, 3597-3607.
- (6) Kabeya, Y. et al. (2000) *EMBO J.* 19, 5720-5728.
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- (9) Wu, J. et al. (2006) *Biochem. Biophys. Res. Commun.* 339, 437-442.
- (10) Ichimura, Y. et al. (2000) *Nature* 408, 488-492.
- (11) Kabeya, Y. et al. (2004) *J. Cell Sci.* 117, 2805-2812.