SANTA CRUZ BIOTECHNOLOGY, INC.

Atg12 siRNA (h): sc-72578

BACKGROUND

Atg12 (autophagy-related protein 12), also known as APG12, APG12L, FBR93 or HAPG12, is a 140 amino acid protein that is ubiquitously expressed and belongs to the Atg12 family of proteins. Atg12 is a homolog of the yeast protein Apg12 that participates in autophagy. Autophagy is a membrane trafficking mechanism that delivers cytoplasmic cargo to the vacuole/lysosome for degradation and recycling. In yeast, autophagy requires a protein conjugation system consisting of Apg12 covalently bound at the carboxy terminal glycine to lysine 149 of Apg5. Similarly in humans, Atg12 is essential for autophagy and localizes to the cytoplasm where it is covalently bound to APG5, a conjugation reaction that requires APG7, Atg10 and ATP. The Atg12-APG5 conjugate functions as an important regulator of the autophagic process and is required for the change in membrane morphology and development of autophagosomes. Due to alternative splicing events, two Atg12 isoforms exist.

REFERENCES

1. Ueno, K., et al. 1998. Cloning and tissue expression of cDNAs from chromosome 5q21-22 which is frequently deleted in advanced lung cancer. Hum. Genet. 102: 63-68.
2. Mizushima, N., et al. 1998. A new protein conjugation system in human. The counterpart of the yeast Apg12p conjugation system essential for autophagy. J. Biol. Chem. 273: 33889-33892.

CHROMOSOMAL LOCATION

Genetic locus: ATG12 (human) mapping to 5q22.3.

PRODUCT

Atg12 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Atg12 shRNA Plasmid (h): sc-72578-SH and Atg12 shRNA (h) Lentiviral Particles: sc-72578-V as alternate gene silencing products.

For independent verification of Atg12 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-72578A, sc-72578B and sc-72578C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Atg12 siRNA (h) is recommended for the inhibition of Atg12 expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

Atg12 (C-6): sc-271688 is recommended as a control antibody for monitoring of Atg12 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Atg12 gene expression knockdown using RT-PCR Primer: Atg12 (h)-PR: sc-72578-PR (20 µl, 477 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Shi, Y., et al. 2012. Autophagy protects against oxaliplatin-induced cell death via ER stress and ROS in Caco-2 cells. PLoS ONE 7: e51076.
2. Wang, Y., et al. 2014. Galangin suppresses Hep G2 cell proliferation by activating the TGF-β receptor/Smad pathway. Toxicology 326: 9-17.
3. Tsai, J.P., et al. 2015. Licochalcone A induces autophagy through PI3K/Akt/mTOR inactivation and autophagy suppression enhances licochalcone A-induced apoptosis of human cervical cancer cells. Oncotarget 6: 28851-28866.
4. Tang, B., et al. 2016. *Fusobacterium nucleatum*-induced impairment of autophagic flux enhances the expression of proinflammatory cytokines via ROS in Caco-2 cells. PLoS ONE 11: e0165701.
5. Li, N., et al. 2017. *Helicobacter pylori* CagA protein negatively regulates autophagy and promotes inflammatory response via c-Met-PI3K/Akt-mTOR signaling pathway. Front. Cell. Infect. Microbiol. 7: 417.
6. Zhu, P., et al. 2017. *Helicobacter pylori* VacA induces autophagic cell death in gastric epithelial cells via the endoplasmic reticulum stress pathway. Cell Death Dis. 8: 3207.
7. Scherr, A.L., et al. 2020. Knockdown of Atg7 induces nuclear-LC3 dependent apoptosis and augments chemotherapy in colorectal cancer cells. Int. J. Mol. Sci. 21: 1099.

RESEARCH USE

For research use only, not for use in diagnostic procedures.