

MAP LC3 α siRNA (h): sc-106197

BACKGROUND

Microtubule-associated proteins (MAPs) regulate microtubule stability and play critical roles in neuronal development and in maintaining the balance between neuronal plasticity and rigidity. MAP light chain 3 β (MAP LC3 β) and MAP light chain 3 α (MAP LC3 α) are subunits that can associate with either MAP-1A or MAP-1B. While MAP LC3 β is essential for autophagy and is associated with autophagosome membranes after processing, MAP LC3 α is involved in the formation of autophagosomal vacuoles and is localized to the intracytoplasmic membrane. MAP LC3 α is expressed as two alternatively spliced isoforms that are expressed in testis, brain, heart, liver and skeletal muscle, but are absent in thymus and peripheral blood leukocytes.

CHROMOSOMAL LOCATION

Genetic locus: MAP1LC3A (human) mapping to 20q11.22.

PRODUCT

MAP LC3 α 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MAP LC3 α shRNA Plasmid (h): sc-106197-SH and MAP LC3 α shRNA (h) Lentiviral Particles: sc-106197-V as alternate gene silencing products.

For independent verification of MAP LC3 α (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-106197A, sc-106197B and sc-106197C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

MAP LC3 α siRNA (h) is recommended for the inhibition of MAP LC3 α 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 μ M in 66 μ l. 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

MAP LC3 α / β (G-4): sc-398822 is recommended as a control antibody for monitoring of MAP LC3 α 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).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor MAP LC3 α gene expression knockdown using RT-PCR Primer: MAP LC3 α (h)-PR: sc-106197-PR (20 μ l, 447 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Blanchet, F.P., et al. 2010. Human immunodeficiency virus-1 inhibition of immunoamphisomes in dendritic cells impairs early innate and adaptive immune responses. *Immunity* 32: 654-669.
2. Chang, C.Y., et al. 2014. Autophagy contributes to gefitinib-induced glioma cell growth inhibition. *Exp. Cell Res.* 327: 102-112.
3. He, P.X., et al. 2014. G226, a novel epipolythiodioxopiperazine derivative, induces autophagy and caspase-dependent apoptosis in human breast cancer cells *in vitro*. *Acta Pharmacol. Sin.* 35: 1055-1064.
4. Wang, Y., et al. 2015. CGK733-induced LC3 II formation is positively associated with the expression of cyclin-dependent kinase inhibitor p21^{Waf1/Cip1} through modulation of the AMPK and PERK/CHOP signaling pathways. *Oncotarget* 6: 39692-39701.
5. Chauhan, S., et al. 2015. Pharmaceutical screen identifies novel target processes for activation of autophagy with a broad translational potential. *Nat. Commun.* 6: 8620.
6. Hou, B., et al. 2019. SQSTM1/p62 loss reverses the inhibitory effect of sunitinib on autophagy independent of AMPK signaling. *Sci. Rep.* 9: 11087.

RESEARCH USE

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

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.