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SANTA CRUZ BIOTECHNOLOGY, INC.

MAP LC3α/β siRNA (m): sc-156052

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-LC3β) and MAP-light chain 3 α (MAP-LC3α) are subunits that can associate with either MAP-1A or MAP-1B. While MAP-LC3β is essential for autophagy and is associated with autophagosome membranes after processing, MAP LC3α is involved in the formation of autophagosomal vacuoles and is localized to the intracytoplasmic membrane. MAP LC3α 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. MAP LC3β, which exists in a cytosolic and a membrane-bound form, may also be involved in formation of autophagosomal vacuoles and is expressed primarily in heart, testis, brain and skeletal muscle.

REFERENCES

1. Fink, J.K., et al. 1996. Human microtubule-associated protein 1A (MAP1A) gene: genomic organization, cDNA sequence, and developmental and tissue-specific expression. Genomics 35: 577-585.
2. Mann, S.S., et al. 1996. Gene localization and developmental expression of light chain 3: a common subunit of microtubule-associated protein 1A (MAP-1A) and MAP-1B. J. Neurosci. Res. 43: 535-544.
3. Zhou, B., et al. 1997. Microtubule-associated protein 1 light chain 3 is a Fibronectin mRNA-binding protein linked to mRNA translation in lamb vascular smooth muscle cells. J. Clin. Invest. 100: 3070-3082.
4. Zhou, B., et al. 1998. Microtubule involvement in translational regulation of Fibronectin expression by light chain 3 of microtubule-associated protein 1 in vascular smooth muscle cells. Circ. Res. 83: 481-489.
5. Kabeya, Y., et al. 2000. LC3, a mammalian homolog of yeast Apg8p, is localized in autophagosome membrane after processing. EMBO J. 19: 5720-5728.
6. Paz, Y., et al. 2000. Structure of GATE-16, membrane transport modulator and mammalian ortholog of autophagocytosis factor Aut7p. J. Biol. Chem. 275: 25445-25450.

CHROMOSOMAL LOCATION

Genetic locus: Map1lc3a (mouse) mapping to 2 H1, Map1lc3b (mouse) mapping to 8 E1.

PRODUCT

MAP LC3α/β siRNA (m) is a pool of 4 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 MAP LC3α/β shRNA Plasmid (m): sc-156052-SH and MAP LC3α/β shRNA (m) Lentiviral Particles: sc-156052-V as alternate gene silencing products.

For independent verification of MAP LC3α/β (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-156052A, sc-156052B, sc-156052C and sc-156052D.

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

MAP LC3α/β siRNA (m) is recommended for the inhibition of MAP LC3α/β expression in mouse 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

MAP LC3α/β (G-4): sc-398822 is recommended as a control antibody for monitoring of MAP LC3α/β 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 LC3α/β gene expression knockdown using RT-PCR Primer: MAP LC3α/β (m)-PR: sc-156052-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

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.