SANTA CRUZ BIOTECHNOLOGY, INC.

Atg14 siRNA (m): sc-142784

BACKGROUND

Atg14 (ATG14 autophagy related 14 homolog), also known as BARKOR (beclin 1-associated autophagy-related key regulator) or ATG14L, is a 492 amino acid protein that contains 3 coiled-coil domains in its N-terminal region. Conserved in chimpanzee, canine, bovine, mouse, rat, chicken, zebrafish, fruit fly and mosquito, Atg14 exhibits highest expression in brain and lowest expression in kidney and ovary. Atg14 localizes to isolation membranes of forming autophagosomes, cytoplasm, endoplasmic reticulum and an uncharacterized punctate structure. Necessary for both basal and inducible autophagy, Atg14 plays a role in autophagosome formation and MAP1LC3/LC3 conjugation to phosphatidylethanolamine. Atg14 promotes BECN1 translocation from the trans-Golgi network to autophagosomes, and forms a complex with BECN1, PI 3-kinase p100 and p150. Atg14 does not form complexes with KIAA0226/Rubicon or UVRAG, which forms a mutually exclusive complex with BECN1 through direct competition with Atg14.

REFERENCES

1. Suzuki, K., et al. 2004. Interrelationships among Atg proteins during autophagy in Saccharomyces cerevisiae. Yeast 21: 1057-1065.
2. Deretic, V. 2006. Autophagy as an immune defense mechanism. Curr. Opin. Immunol. 18: 375-382.
3. Liang, C., et al. 2006. Autophagic and tumour suppressor activity of a novel Beclin1-binding protein UVRAG. Nat. Cell Biol. 8: 688-699.
4. Xie, Z. and Klionsky, D.J. 2007. Autophagosome formation: core machinery and adaptations. Nat. Cell Biol. 9: 1102-1109.
5. Mari, M. and Reggiori, F. 2007. Shaping membranes into autophagosomes. Nat. Cell Biol. 9: 1125-1127.
6. Itakura, E., et al. 2008. Beclin 1 forms two distinct phosphatidylinositol 3-kinase complexes with mammalian Atg14 and UVRAG. Mol. Biol. Cell 19: 5360-5372.

CHROMOSOMAL LOCATION

Genetic locus: Atg14 (mouse) mapping to 14 C1.

PRODUCT

Atg14 siRNA (m) 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 Atg14 shRNA Plasmid (m): sc-142784-SH and Atg14 shRNA (m) Lentiviral Particles: sc-142784-V as alternate gene silencing products.

For independent verification of Atg14 (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-142784A, sc-142784B and sc-142784C.

PROTOCOLS

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

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

Atg14 siRNA (m) is recommended for the inhibition of Atg14 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Atg14 gene expression knockdown using RT-PCR Primer: Atg14 (m)-PR: sc-142784-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.