SANTA CRUZ BIOTECHNOLOGY, INC.

Rubicon siRNA (h): sc-78326

BACKGROUND

Rubicon, also known as KIAA0226 or Baron (Beclin-1 associated RUN domain containing protein), is a 972 amino acid protein that is responsible for negatively regulating endocytic trafficking. Rubicon impairs the maturation of the autophagosome and may compromise VPS34 activity. Rubicon is found in the early/late endosome and the lysosome. Rubicon forms a complex with BECN1, VPS34, p150 and UVRAG, which down-regulates autophagy. Mutation in the gene encoding Rubicon may lead to a new form of recessive ataxia called Salih ataxia, which is an early childhood-onset and may be characterized by epilepsy and mental retardation. Rubicon exists as three alternatively spliced isoforms and is post-translationally phosphorylated on multiple serine and threonine residues. The gene encoding Rubicon maps to chromosome 3, which consists of about 214 million bases encoding over 1,100 genes.

REFERENCES

1. Nagase, T., et al. 1996. Prediction of the coding sequences of unidentified human genes. VI. The coding sequences of 80 new genes (KIAA0201-KIAA0280) deduced by analysis of cDNA clones from cell line KG-1 and brain. DNA Res. 3: 321-329, 341-354.
2. Matsunaga, K., et al. 2009. Binding Rubicon to cross the Rubicon. Autophagy 5: 876-877.
3. Matsunaga, K., et al. 2009. Two Beclin 1-binding proteins, Atg14L and Rubicon, reciprocally regulate autophagy at different stages. Nat. Cell Biol. 11: 385-396.
4. Assoum, M., et al. 2010. Rundataxin, a novel protein with RUN and diacylglycerol binding domains, is mutant in a new recessive ataxia. Brain 133: 2439-2447.
5. Tabata, K., et al. 2010. Rubicon and PLEKHM1 negatively regulate the endocytic/autophagic pathway via a novel Rab7-binding domain. Mol. Biol. Cell 21: 4162-4172.
6. Sun, Q., et al. 2010. Rubicon controls endosome maturation as a Rab7 effector. Proc. Natl. Acad. Sci. USA 107: 19338-19343.
7. Yang, C.S., et al. 2012. The autophagy regulator Rubicon is a feedback inhibitor of CARD9-mediated host innate immunity. Cell Host Microbe 11: 277-289.

CHROMOSOMAL LOCATION

Genetic locus: KIAA0226 (human) mapping to 3q29.

PRODUCT

Rubicon 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 Rubicon shRNA Plasmid (h): sc-78326-SH and Rubicon shRNA (h) Lentiviral Particles: sc-78326-V as alternate gene silencing products.

For independent verification of Rubicon (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-78326A, sc-78326B and sc-78326C.

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

Rubicon siRNA (h) is recommended for the inhibition of Rubicon 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Rubicon gene expression knockdown using RT-PCR Primer: Rubicon (h)-PR: sc-78326-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Nanni, M., et al. 2018. Interplay between FGFR2b-induced autophagy and phagocytosis: role of PLCγ-mediated signalling. J. Cell. Mol. Med. 22: 668-683.
2. Kumar, V., et al. 2019. Disrupted apolipoprotein L1-miR193a axis dedifferentiates podocytes through autophagy blockade in an APOL1 risk milieu. Am. J. Physiol., Cell Physiol. 317: C209-C225.
3. Bhattacharya, A., et al. 2023. A lysosome membrane regeneration pathway depends on TBC1D15 and autophagic lysosomal reformation proteins. Nat. Cell Biol. 25: 685-698.

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.