

Rubicon siRNA (m): sc-146442

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.

CHROMOSOMAL LOCATION

Genetic locus: 1700021K19Rik (mouse) mapping to 16 B3.

PRODUCT

Rubicon 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Rubicon shRNA Plasmid (m): sc-146442-SH and Rubicon shRNA (m) Lentiviral Particles: sc-146442-V as alternate gene silencing products.

For independent verification of Rubicon (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-146442A, sc-146442B and sc-146442C.

RESEARCH USE

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

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

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

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Rubicon gene expression knockdown using RT-PCR Primer: Rubicon (m)-PR: sc-146442-PR (20 μ l, 517 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Huang, J.H., et al. 2018. NLRX1 facilitates *Histoplasma capsulatum*-induced LC3-associated phagocytosis for cytokine production in macrophages. *Front. Immunol.* 9: 2761.
2. Li, Y., et al. 2020. Podocyte EGFR inhibits autophagy through upregulation of Rubicon in type II diabetic nephropathy. *Diabetes*. E-published.

PROTOCOLS

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