SANTA CRUZ BIOTECHNOLOGY, INC.

RIP siRNA (h): sc-36426

BACKGROUND

In contrast to growth factors which promote cell proliferation, FAS ligand (FAS-L) and the tumor necrosis factors (TNFs) rapidly induce apoptosis. Cellular response to FAS-L and TNF is mediated by structurally related receptors containing a conserved "death domain" and belonging to the TNF receptor superfamily. TRADD, FADD and RIP are FAS/TNF-R1 interacting proteins that contain a death domain homologous region (DDH). TRADD (TNF-R1-associated death domain) and FADD (FAS-associated death domain) associate with the death domains of both FAS and TNF-R1 via their DDH regions. Overexpression of TRADD leads to NFkB activation and apoptosis in the absence of TNF. Overexpression of FADD causes apoptosis, which can be blocked by the cow pox protein CrmA, suggesting that FADD lies upstream of ICE and possibly other serine proteases. The receptor interacting protein, RIP, associates with FAS exclusively via its DDH and this association is abrogated in lpr mutants. Unlike TRADD and FADD, RIP contains a putative amino terminal kinase domain.

REFERENCES

1. Smith, C.A., et al. 1994. The TNF receptor superfamily of cellular and viral proteins: activation, costimulation and death. Cell 76: 959-962.
2. Nagata, S., et al. 1995. The FAS death factor. Science 267: 1449-1456.
3. Sato, T., et al. 1995. FAP-1: a protein tyrosine phosphatase that associates with FAS. Science 268: 411-414.

CHROMOSOMAL LOCATION

Genetic locus: RIPK1 (human) mapping to 6p25.2.

PRODUCT

RIP 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 RIP shRNA Plasmid (h): sc-36426-SH and RIP shRNA (h) Lentiviral Particles: sc-36426-V as alternate gene silencing products.

For independent verification of RIP (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-36426A, sc-36426B and sc-36426C.

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

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

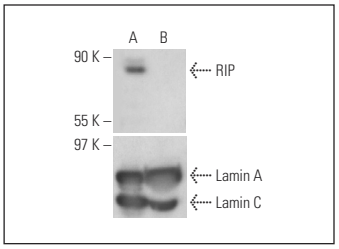
GENE EXPRESSION MONITORING

RIP (C-12): sc-133102 is recommended as a control antibody for monitoring of RIP 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor RIP gene expression knockdown using RT-PCR Primer: RIP (h)-PR: sc-36426-PR (20 µl, 438 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

DATA



RIP siRNA (h): sc-36426. Western blot analysis of RIP expression in non-transfected control (**A**) and RIP siRNA transfected (**B**) HeLa cells. Blot probed with RIP (K-20): sc-1169. Lamin A/C (H-110): sc-20681 used as specificity and loading control.

SELECT PRODUCT CITATIONS

1. Hunter, I. and Nixon, G.F. 2006. Spatial compartmentalization of tumor necrosis factor (TNF) receptor 1-dependent signaling pathways in human airway smooth muscle cells lipid rafts are essential for TNF-α-mediated activation of RhoA but dispensable for the activation of the NFκB and mapk pathways. J. Biol. Chem. 281: 34705-34715.
2. Funakoshi, T., et al. 2016. Necroptosis-like neuronal cell death caused by cellular cholesterol accumulation. J. Biol. Chem. 291: 25050-25065.
3. Lim, R., et al. 2017. TLR2, TLR3 and TLR5 regulation of pro-inflammatory and pro-labour mediators in human primary myometrial cells. J. Reprod. Immunol. 122: 28-36.

RESEARCH USE

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