

# FADD siRNA (h): sc-35352

## BACKGROUND

In contrast to growth factors which promote cell proliferation, FAS ligand (FAS-L) and tumor necrosis factor (TNF) 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-RI interacting proteins that contain a death domain homologous region (DDH). TRADD (TNF-RI-associated death domain) and FADD (FAS-associated death domain) associate with the death domains of both FAS and TNF-RI via their DDH regions. Overexpression of TRADD leads to NF $\kappa$ B activation and apoptosis in the absence of TNF. Overexpression of FADD causes apoptosis, which can be blocked by the bovine 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.

## CHROMOSOMAL LOCATION

Genetic locus: FADD (human) mapping to 11q13.3.

## PRODUCT

FADD siRNA (h) is a target-specific 19-25 nt siRNA designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FADD shRNA Plasmid (h): sc-35352-SH and FADD shRNA (h) Lentiviral Particles: sc-35352-V as alternate gene silencing 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

FADD siRNA (h) is recommended for the inhibition of FADD 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  $\mu$ M in 66  $\mu$ 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.

## GENE EXPRESSION MONITORING

FADD (G-4): sc-271748 is recommended as a control antibody for monitoring of FADD 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 FADD gene expression knockdown using RT-PCR Primer: FADD (h)-PR: sc-35352-PR (20  $\mu$ l, 491 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Garnett, T.O., et al. 2006. Accelerated degradation of FADD and procaspase 8 in cells expressing human papilloma virus 16 E6 impairs TRAIL-mediated apoptosis. *Cell Death Differ.* 13: 1915-1926.
2. Lai, Y.M., et al. 2007. Induction of cell cycle arrest and apoptosis by BCG infection in cultured human bronchial airway epithelial cells. *Am. J. Physiol. Lung Cell. Mol. Physiol.* 293: L393-L401.
3. Liu, W.H., et al. 2009. ROS-mediated p38 $\alpha$  MAPK activation and ERK inactivation responsible for upregulation of Fas and FasL and autocrine Fas-mediated cell death in Taiwan cobra phospholipase A<sub>2</sub>-treated U937 cells. *J. Cell. Physiol.* 219: 642-651.
4. Liu, W.H., et al. 2010. Piceatannol induces Fas and FasL up-regulation in human leukemia U937 cells via Ca<sup>2+</sup>/p38 $\alpha$  MAPK-mediated activation of c-Jun and ATF-2 pathways. *Int. J. Biochem. Cell Biol.* 42: 1498-1506.
5. Liu, W.H. and Chang, L.S. 2011. Adaphostin promotes caffeine-evoked autocrine Fas-mediated death pathway activation in Bcr/Abl-positive leukaemia cells. *Biochem. J.* 439: 453-467.
6. Liu, W.H., et al. 2013. P38 MAPK/PP2A $\alpha$ /TTP pathway on the connection of TNF- $\alpha$  and caspases activation on hydroquinone-induced apoptosis. *Carcinogenesis* 34: 818-827.
7. Huang, C.H., et al. 2016. The association between p38 MAPK-mediated TNF- $\alpha$ /TNFR2 up-regulation and 2-(4-aminophenyl)-7-methoxybenzothiazole-induced apoptosis in human leukemia U937 cells. *J. Cell. Physiol.* 231: 130-141.
8. Helmke, C., et al. 2016. Ligand stimulation of CD95 induces activation of PIK3 followed by phosphorylation of caspase-8. *Cell Res.* 26: 914-934.
9. Wang, S.W., et al. 2018. Stimulation of Fas/FasL-mediated apoptosis by luteolin through enhancement of Histone H3 acetylation and c-Jun activation in HL-60 leukemia cells. *Mol. Carcinog.* 57: 866-877.
10. Wang, L.J., et al. 2019. Non-mitotic effect of albendazole triggers apoptosis of human leukemia cells via SIRT3/ROS/p38 MAPK/TTP axis-mediated TNF- $\alpha$  upregulation. *Biochem. Pharmacol.* 162: 154-168.

## RESEARCH USE

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