

## FLIP<sub>S/L</sub> siRNA (m): sc-35389

### BACKGROUND

FLIP (FLICE inhibitory protein) is expressed as both long and short forms and is involved in the regulation of apoptosis. The short form of FLIP contains two death effector domains homologous to the death effector domain of the Fas-associating protein FADD. The long form of FLIP, which shares significant homology with the cysteine protease FLICE, contains an additional caspase-like domain, but lacks a catalytic active site and lacks the residues that form the substrate binding pocket in most caspases. FLIP has been designated by independent groups as Casper, I-FLICE, CLARP, FLAME-1 and MRIT. Although its exact role is still being elucidated, FLIP appears to be an important factor in the regulation of apoptosis downstream of all known death receptors.

### REFERENCES

1. Thome, M., et al. 1997. Viral FLICE-inhibitory proteins (FLIPs) prevent apoptosis induced by death receptors. *Nature* 386: 517-521.
2. Imler, M., et al. 1997. Inhibition of death receptor signals by cellular FLIP. *Nature* 388: 190-195.
3. Shu, H.B., et al. 1997. Casper is a FADD- and caspase-related inducer of apoptosis. *Immunity* 6: 751-763.
4. Hu, S., et al. 1997. I-FLICE, a novel inhibitor of tumor necrosis factor receptor-1 and CD95-induced apoptosis. *J. Biol. Chem.* 272: 17255-17257.
5. Srinivasula, S.M., et al. 1997. FLAME-1, a novel FADD-like anti-apoptotic molecule that regulates Fas/TNFR1-induced apoptosis. *J. Biol. Chem.* 272: 18542-18545.

### CHROMOSOMAL LOCATION

Genetic locus: Cflar (mouse) mapping to 1 C1.3.

### PRODUCT

FLIP<sub>S/L</sub> siRNA (m) is a pool of 4 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see FLIP<sub>S/L</sub> shRNA Plasmid (m): sc-35389-SH and FLIP<sub>S/L</sub> shRNA (m) Lentiviral Particles: sc-35389-V as alternate gene silencing products.

For independent verification of FLIP<sub>S/L</sub> (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-35389A, sc-35389B, sc-35389C and sc-35389D.

### 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

FLIP<sub>S/L</sub> siRNA (m) is recommended for the inhibition of FLIP<sub>S/L</sub> 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  $\mu$ 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

FLIP<sub>S/L</sub> (G-11): sc-5276 is recommended as a control antibody for monitoring of FLIP<sub>S/L</sub> 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).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

### RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor FLIP<sub>S/L</sub> gene expression knockdown using RT-PCR Primer: FLIP<sub>S/L</sub> (m)-PR: sc-35389-PR (20  $\mu$ l, 372 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

### SELECT PRODUCT CITATIONS

1. Chandrasekaran, Y., et al. 2006. Influence of TRP53 status on Fas membrane localization, CFLAR (c-FLIP) ubiquitinylation, and sensitivity of GC-2spd (ts) cells to undergo Fas-mediated apoptosis. *Biol. Reprod.* 74: 560-568.
2. Wang, D., et al. 2013. The histone deacetylase inhibitor vorinostat prevents TNF $\alpha$ -induced necroptosis by regulating multiple signaling pathways. *Apoptosis* 18: 1348-1362.
3. Tenshin, H., et al. 2017. TAK1 inhibition subverts the osteoclastogenic action of TRAIL while potentiating its antimyeloma effects. *Blood Adv.* 1: 2124-2137.

### RESEARCH USE

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