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SANTA CRUZ BIOTECHNOLOGY, INC.

FLASH siRNA (h): sc-43761

BACKGROUND

Fas is a member of the tumor necrosis factor family of membrane receptors, which induces apoptosis by binding to its ligand, Fas-L. Fas mediates apoptosis through a group of proteins that bind to its intracellular "death" domain, including FADD. After binding to Fas, FADD binds to caspase-8, resulting in activation of caspase-8 and the initiation of the caspase-mediated apoptotic pathway. FLASH, for FLICE- associated huge protein, has been identified as an additional component of the Fas-FADD-caspase-8 complex, also referred to as the DISC complex. FLASH shares homology with the *C. elegans* CED-4 protein and the mammalian Apaf-1 protein, which are both involved in activating caspases. FLASH was shown to be required for activation of caspase-8 during Fas-mediated apoptosis.

REFERENCES

1. Itoh, N., et al. 1991. The polypeptide encoded by the cDNA for human cell surface antigen Fas can mediate apoptosis. Cell 66: 233-243.
2. Suda, T., et al. 1993. Molecular cloning and expression of the Fas ligand, a novel member of the tumor necrosis factor family. Cell 75: 1169-1178.
3. Chinnaiyan, A.M., et al. 1995. FADD, a novel death domain-containing protein, interacts with the death domain of Fas and initiates apoptosis. Cell 81: 505-512.
4. Boldin, M.P., et al. 1995. A novel protein that interacts with the death domain of Fas/APO1 contains a sequence motif related to the death domain. J. Biol. Chem. 270: 7795-7798.
5. Boldin, M.P., et al. 1996. Involvement of MACH, a novel MORT1/FADD-interacting protease, in Fas/APO-1- and TNF receptor-induced cell death. Cell 85: 803-815.
6. Muzio, M., et al. 1996. FLICE, a novel FADD-homologous ICE/CED-3-like protease, is recruited to the CD95 (Fas/APO-1) death-inducing signaling complex. Cell 85: 817-827.

CHROMOSOMAL LOCATION

Genetic locus: CASP8AP2 (human) mapping to 6q15.

PRODUCT

FLASH 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 FLASH shRNA Plasmid (h): sc-43761-SH and FLASH shRNA (h) Lentiviral Particles: sc-43761-V as alternate gene silencing products.

For independent verification of FLASH (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-43761A, sc-43761B and sc-43761C.

PROTOCOLS

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

FLASH siRNA (h) is recommended for the inhibition of FLASH 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 FLASH gene expression knockdown using RT-PCR Primer: FLASH (h)-PR: sc-43761-PR (20 µl, 455 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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