SANTA CRUZ BIOTECHNOLOGY, INC.

DAPK siRNA (h): sc-38976

BACKGROUND

DAP (death associated protein) kinase and ZIP kinase are members of a novel protein kinase family, the members of which have the capacity to mediate apoptosis through their catalytic activities. DAP kinase (DAPK) contains a "death domain" and has been shown to mediate IFN-γ-induced apoptosis. The introduction of DAPK into highly metastatic carcinoma clones lacking DAPK expression has been shown to result in the suppression of metastasis, thus linking suppression of apoptosis to metastasis. ZIP kinase contains a leucine zipper domain, which is necessary for homodimerization and for interaction with other leucine zipper proteins. ZIP kinase dimerizes with ATF-4, an ATF/CREB transcription factor family member that contains a leucine zipper. Overexpression of ZIP kinase has been shown to result in morphological changes associated with apoptosis in NIH/3T3 cells.

REFERENCES

1. Feinstein, E., et al. 1995. Assignment of DAP1 and DAPK—genes that positively mediate programmed cell death triggered by IFN-γ—to chromosome regions 5p12.2 and 9q34.1, respectively. Genomics 29: 305-307.
2. Sakagami, H., et al. 1997. Molecular cloning and developmental expression of a rat homologue of death-associated protein kinase in the nervous system. Brain Res. Mol. Brain Res. 52: 249-256.
3. Inbal, B., et al. 1997. DAP kinase links the control of apoptosis to metastasis. Nature 390: 180-184.
4. Kawai, T., et al. 1998. ZIP kinase, a novel serine/threonine kinase which mediates apoptosis. Mol. Cell. Biol. 18: 1642-1651.

CHROMOSOMAL LOCATION

Genetic locus: DAPK1 (human) mapping to 9q21.33.

PRODUCT

DAPK 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 DAPK shRNA Plasmid (h): sc-38976-SH and DAPK shRNA (h) Lentiviral Particles: sc-38976-V as alternate gene silencing products.

For independent verification of DAPK (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-38976A, sc-38976B and sc-38976C.

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

DAPK siRNA (h) is recommended for the inhibition of DAPK 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

DAPK (17): sc-136286 is recommended as a control antibody for monitoring of DAPK 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-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor DAPK gene expression knockdown using RT-PCR Primer: DAPK (h)-PR: sc-38976-PR (20 µl, 562 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Rennier, K. and Ji, J.Y. 2012. Shear stress regulates expression of death-associated protein kinase in suppressing TNFα-induced endothelial apoptosis. J. Cell. Physiol. 227: 2398-2411.
2. Arif, A., et al. 2012. Heterotrimeric GAIT complex drives transcript-selective translation inhibition in murine macrophages. Mol. Cell. Biol. 32: 5046-5055.
3. Zhang, H.T., et al. 2014. Hepatitis B virus x protein induces autophagy via activating death-associated protein kinase. J. Viral Hepat. 21: 642-649.

RESEARCH USE

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