SANTA CRUZ BIOTECHNOLOGY, INC.

DDEFL1 siRNA (h): sc-88264

BACKGROUND

DDEFL1 (development and differentiation enhancing factor-like 1), also known as ACAP4, UPLC1, CENTB6 or ASAP3 (ArfGAP with SH3 domain, ankyrin repeat and PH domain 3), is a 903 amino acid cytoplasmic protein belonging to the subfamily of ADP-ribosylation factor (Arf) GTPase-activating proteins. DDEFL1 contains two ANK repeats, an Arf-GAP domain and a PH domain, and is expressed in lung, liver, blood leukocytes and primary hepatocarcinoma. The Arf-GAP domain of DDEFL1 catalyzes the hydrolysis of GTP bound to Arf proteins. DDEFL1 promotes cell differentiation and migration, and has been implicated in the pathogenesis of hepatocellular carcinoma. Existing as two isoforms produced by alternative splicing events, DDEFL1 is encoded by a gene located on human chromosome 1. Human chromosome 1 spans 260 million base pairs, contains over 3,000 genes and comprises nearly 8% of the human genome.

REFERENCES

1. Jackson, T.R., et al. 2000. ACAPs are Arf6 GTPase-activating proteins that function in the cell periphery. J. Cell Biol. 151: 627-638.
2. Randazzo, P.A., et al. 2000. Molecular aspects of the cellular activities of ADP-ribosylation factors. Sci. STKE 2000: re1.
3. Okabe, H., et al. 2004. Isolation of development and differentiation enhancing factor-like 1 (DDEFL1) as a drug target for hepatocellular carcinomas. Int. J. Oncol. 24: 43-48.
4. Randazzo, P.A., et al. 2004. Arf GAPs: multifunctional proteins that regulate membrane traffic and Actin remodelling. Cell. Signal. 16: 401-413.
5. Sabe, H., et al. 2006. ArfGAP family proteins in cell adhesion, migration and tumor invasion. Curr. Opin. Cell Biol. 18: 558-564.
6. Fang, Z., et al. 2006. Proteomic identification and functional characterization of a novel Arf6 GTPase-activating protein, ACAP4. Mol. Cell. Proteomics 5: 1437-1449.
7. Randazzo, P.A., et al. 2007. Arf GAPs as regulators of the Actin cytoskeleton. Biol. Cell 99: 583-600.
8. Inoue, H., et al. 2007. Arf GAPs and their interacting proteins. Traffic 8: 1465-1475.

CHROMOSOMAL LOCATION

Genetic locus: ASAP3 (human) mapping to 1p36.12.

PRODUCT

DDEFL1 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 DDEFL1 shRNA Plasmid (h): sc-88264-SH and DDEFL1 shRNA (h) Lentiviral Particles: sc-88264-V as alternate gene silencing products.

For independent verification of DDEFL1 (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-88264A, sc-88264B and sc-88264C.

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

DDEFL1 siRNA (h) is recommended for the inhibition of DDEFL1 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

DDEFL1 (C-3): sc-514132 is recommended as a control antibody for monitoring of DDEFL1 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 DDEFL1 gene expression knockdown using RT-PCR Primer: DDEFL1 (h)-PR: sc-88264-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Mao, X., et al. 2017. DDEFL1 correlated with Rho GTPases activity in breast cancer. Oncotarget 8: 112487-112497.

RESEARCH USE

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