SANTA CRUZ BIOTECHNOLOGY, INC.

NSFL1C p47 siRNA (h): sc-76032

BACKGROUND

NSFL1C p47, also known as p47, NSFL1C, UBX1, UBXD10 or UBXN2C, is a 370 amino acid protein that localizes to both the nucleus and the Golgi apparatus (specifically to Golgi stacks) and contains one SEP domain and one UBX domain. Functioning as part of a ternary complex with VCP (a protein involved in the heterotypic fusion of transport vesicles with their target membranes) and Syntaxin 5, NSFL1C p47 interacts with and reduces the ATPase activity of VCP and is required for the fragmentation of Golgi stacks during mitosis and for subsequent reassembly of Golgi stacks after mitosis. NSFL1C p47 is subject to phosphorylation during mitosis, which inhibits NSFL1C p47-Golgi interaction and is, therefore, required for proper Golgi stack formation and cisternal regrowth. Human NSFL1C p47 shares 89% sequence identity with its mouse counterpart, suggesting a conserved role between species. Multiple isoforms of NSFL1C p47 exist due to alternative splicing events.

REFERENCES

1. Kondo, H., et al. 1997. p47 is a cofactor for p97-mediated membrane fusion. Nature 388: 75-78.
2. Rabouille, C., et al. 1998. Syntaxin 5 is a common component of the NSF- and p97-mediated reassembly pathways of Golgi cisternae from mitotic Golgi fragments *in vitro*. Cell 92: 603-610.
3. Ye, Y., et al. 2001. The AAA ATPase Cdc48/p97 and its partners transport proteins from the ER into the cytosol. Nature 414: 652-656.
4. Meyer, H.H., et al. 2002. Direct binding of ubiquitin conjugates by the mammalian p97 adaptor complexes, p47 and Ufd1-Npl4. EMBO J. 21: 5645-5652.

CHROMOSOMAL LOCATION

Genetic locus: NSFL1C (human) mapping to 20p13.

PRODUCT

NSFL1C p47 siRNA (h) is a pool of 2 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 NSFL1C p47 shRNA Plasmid (h): sc-76032-SH and NSFL1C p47 shRNA (h) Lentiviral Particles: sc-76032-V as alternate gene silencing products.

For independent verification of NSFL1C p47 (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-76032A and sc-76032B.

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

NSFL1C p47 siRNA (h) is recommended for the inhibition of NSFL1C p47 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

NSFL1C p47 (D-9): sc-365215 is recommended as a control antibody for monitoring of NSFL1C p47 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 NSFL1C p47 gene expression knockdown using RT-PCR Primer: NSFL1C p47 (h)-PR: sc-76032-PR (20 µl, 597 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Gonsalves, C.S., et al. 2014. Angiogenic growth factors augment K—Cl cotransporter expression in erythroid cells via hypoxia-inducible factor-1α. Am. J. Hematol. 89: 273-281.
2. Lin, C.P., et al. 2016. Simvastatin attenuates oxidative stress, NFκB activation, and artery calcification in LDLR$^{-/-}$ mice fed with high fat diet via down-regulation of tumor necrosis factor-α and TNF receptor 1. PLoS ONE 10: e0143686.
3. Chang, T.T., et al. 2020. Hydralazine improves ischemia-induced neovasculogenesis via xanthine-oxidase inhibition in chronic renal insufficiency. Pharmacol. Res. 151: 104509.

RESEARCH USE

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