SANTA CRUZ BIOTECHNOLOGY, INC.

NEDD4 siRNA (h): sc-41079

BACKGROUND

NEDD4 family interacting protein-1 (NDFIP1), also known as N4WBP5, is a member of a family of highly conserved proteins. NEDD4 is a 221 amino acid, Golgi-associated protein which contains three transmembrane domains in its carboxy-terminus and two PY motifs in its amino-terminus. NEDD4 may play a role in Golgi structure and function. It binds the WW domains of a number of NEDD4 family members called HECT-type E3 ubiquitin ligases. NEDD4 is strongly expressed in surviving neurons around an injury site, which suggests that ubiquitination may be a possible survival strategy and that NEDD4 may act as a neuroprotective protein. It may also have an effect on the function of Itch, another E3 ubiquitin ligase. Expression of NEDD4 and its association with Itch may be promoted by T cell activation. Mice lacking NEDD4 exhibit inactivation of Itch and accumulation of Jun B, causing severe skin and lung inflammation and premature death.

REFERENCES

1. Harvey, K.F., et al. 2002. N4WBP5, a potential target for ubiquitination by the NEDD4 family of proteins, is a novel Golgi-associated protein. J. Biol. Chem. 277: 9307-9317.
2. Donnison, M., et al. 2004. Isolation of genes associated with developmentally competent bovine oocytes and quantitation of their levels during development. Biol. Reprod. 71: 1813-1821.

CHROMOSOMAL LOCATION

Genetic locus: NEDD4 (human) mapping to 15q21.3.

PRODUCT

NEDD4 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 NEDD4 shRNA Plasmid (h): sc-41079-SH and NEDD4 shRNA (h) Lentiviral Particles: sc-41079-V as alternate gene silencing products.

For independent verification of NEDD4 (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-41079A, sc-41079B and sc-41079C.

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

NEDD4 siRNA (h) is recommended for the inhibition of NEDD4 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

NEDD4 (F-11): sc-518160 is recommended as a control antibody for monitoring of NEDD4 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NEDD4 gene expression knockdown using RT-PCR Primer: NEDD4 (h)-PR: sc-41079-PR (20 µl, 511 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Kim, S.M., et al. 2010. Acquired resistance to cetuximab is mediated by increased PTEN instability and leads cross-resistance to gefitinib in HCC827 NSCLC cells. Cancer Lett. 296: 150-159.
2. Klein, B.Y., et al. 2011. PI3K/Akt responses to oxytocin stimulation in Caco2BB gut cells. J. Cell. Biochem. 112: 3216-3226.
3. Sugeno, N., et al. 2014. Lys-63-linked ubiquitination by E3 ubiquitin ligase NEDD4-1 facilitates endosomal sequestration of internalized α-synuclein. J. Biol. Chem. 289: 18137-18151.
4. Lin, A. and Man, H.Y. 2014. Endocytic adaptor epidermal growth factor receptor substrate 15 (Eps15) is involved in the trafficking of ubiquitinated α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors. J. Biol. Chem. 289: 24652-24664.
5. Xie, P., et al. 2021. Neddylation of PTEN regulates its nuclear import and promotes tumor development. Cell Res. 31: 291-311.
6. Cid-Díaz, T., et al. 2021. Obestatin signalling counteracts glucocorticoid-induced skeletal muscle atrophy via NEDD4/KLF15 axis. J. Cachexia Sarcopenia Muscle 2: 493-505.

RESEARCH USE

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

PROTOCOLS

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