UBE2NL siRNA (h): sc-106660



The Power to Question

BACKGROUND

Ubiquitination is an important mechanism through which three classes of enzymes act in concert to target short-lived or abnormal proteins for destruction. The three classes of enzymes involved in ubiquitination are the ubiquitinactivating enzymes (E1s), the ubiquitin-conjugating enzymes (E2s) and the ubiquitin-protein ligases (E3s). UBE2NL (ubiquitin-conjugating enzyme E2N-like) is a 153 amino acid protein that belongs to the ubiquitin-conjugating enzyme family and may have similar functions as UBE2N. UBE2N, also known as UBC13, mediates the transcription of several target genes and is thought to play a role in cell cycle progression, cellular differentiation and DNA repair mechanisms that ensure cell survival after DNA damage. UBE2NL is encoded by a gene that is located on human chromosome X, which consists of about 153 million base pairs and nearly 1,000 genes.

REFERENCES

- Yamaguchi, T., Kim, N.S., Sekine, S., Seino, H., Osaka, F., Yamao, F. and Kato, S. 1996. Cloning and expression of cDNA encoding a human ubiquitinconjugating enzyme similar to the *Drosophila* bendless gene product. J. Biochem. 120: 494-497.
- Hoege, C., Pfander, B., Moldovan, G.L., Pyrowolakis, G. and Jentsch, S. 2002. RAD6-dependent DNA repair is linked to modification of PCNA by ubiquitin and SUMO. Nature 419: 135-141.
- 3. Andersen, P.L., Zhou, H., Pastushok, L., Moraes, T., McKenna, S., Ziola, B., Ellison, M.J., Dixit, V.M. and Xiao, W. 2005. Distinct regulation of Ubc13 functions by the two ubiquitin-conjugating enzyme variants Mms2 and Uev1A. J. Cell Biol. 170: 745-755.
- Plans, V., Scheper, J., Soler, M., Loukili, N., Okano, Y. and Thomson, T.M. 2006. The RING finger protein RNF8 recruits UBC13 for lysine 63-based self polyubiquitylation. J. Cell. Biochem. 97: 572-582.
- Yamamoto, M., Okamoto, T., Takeda, K., Sato, S., Sanjo, H., Uematsu, S., Saitoh, T., Yamamoto, N., Sakurai, H., Ishii, K.J., Yamaoka, S., Kawai, T., Matsuura, Y., Takeuchi, O. and Akira, S. 2006. Key function for the Ubc13 E2 ubiquitin-conjugating enzyme in immune receptor signaling. Nat. Immunol. 7: 962-970.

CHROMOSOMAL LOCATION

Genetic locus: UBE2NL (human) mapping to Xq27.3.

PRODUCT

UBE2NL 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see UBE2NL shRNA Plasmid (h): sc-106660-SH and UBE2NL shRNA (h) Lentiviral Particles: sc-106660-V as alternate gene silencing products.

For independent verification of UBE2NL (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-106660A, sc-106660B and sc-106660C.

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 μ l of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNAse-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

UBE2NL siRNA (h) is recommended for the inhibition of UBE2NL 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

UBC13/UBE2NL (C-9): sc-365722 is recommended as a control antibody for monitoring of UBE2NL 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG κ 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 UBE2NL gene expression knockdown using RT-PCR Primer: UBE2NL (h)-PR: sc-106660-PR (20 μ l). 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.

PROTOCOLS

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