

UBE2E3 siRNA (m): sc-61749

BACKGROUND

The ubiquitin (Ub) pathway involves three sequential enzymatic steps that facilitate the conjugation of Ub and Ub-like molecules to specific protein substrates. The first step requires the ATP-dependent activation of the Ub C-terminus and the assembly of multi-Ub chains by the Ub-activating enzyme known as the E1 component. The Ub chain is then conjugated to the Ub-conjugating enzyme (E2) to generate an intermediate Ub-E2 complex. The Ub-ligase (E3) then catalyzes the transfer of Ub from E2 to the appropriate protein substrate. A wide range of enzymes facilitate in the proteolytic Ub pathway including UBE2E3, also designated UBCH9, which catalyzes the covalent attachment of ubiquitin to other proteins and is involved in the regulation of transepithelial sodium transport in renal cells. UBE2E3 may also be involved in cell growth arrest. The UBE2E3 protein shuttles between the cytoplasm and nucleus in a IPO11-dependent manner. It is ubiquitously expressed at low levels and is highly expressed in skeletal muscle.

REFERENCES

1. Woods, Y.L., Xirodimas, D.P., Prescott, A.R., Sparks, A., Lane, D.P. and Saville, M.K. 2004. p14 Arf promotes small ubiquitin-like modifier conjugation of Werners helicase. *J. Biol. Chem.* 279: 50157-50166.
2. Debonneville, C. and Staub, O. 2004. Participation of the ubiquitin-conjugating enzyme UBE2E3 in Nedd4-2-dependent regulation of the epithelial Na⁺ channel. *Mol. Cell. Biol.* 24: 2397-2409.
3. Hsu, Y.J., Zimmer, W.E. and Goodman, S.R. 2005. Erythrocyte spectrin's chimeric E2/E3 ubiquitin conjugating/ligating activity. *Cell. Mol. Biol.* 51: 187-193.
4. Chang, T.L., Kakhniashvili, D.G. and Goodman, S.R. 2005. Spectrin's E2/E3 ubiquitin conjugating/ligating activity is diminished in sickle cells. *Am. J. Hematol.* 79: 89-96.
5. Wang, W., Attia, A.S., Liu, L., Rosche, T., Wagner, N.J. and Hansen, E.J. 2005. Development of a shuttle vector for *Moraxella catarrhalis*. *Plasmid* 55: 50-57.

CHROMOSOMAL LOCATION

Genetic locus: Ube2e3 (mouse) mapping to 2 C3.

PRODUCT

UBE2E3 siRNA (m) 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 UBE2E3 shRNA Plasmid (m): sc-61749-SH and UBE2E3 shRNA (m) Lentiviral Particles: sc-61749-V as alternate gene silencing products.

For independent verification of UBE2E3 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-61749A, sc-61749B and sc-61749C.

PROTOCOLS

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

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

UBE2E3 siRNA (m) is recommended for the inhibition of UBE2E3 expression in mouse 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 μ M in 66 μ l. 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor UBE2E3 gene expression knockdown using RT-PCR Primer: UBE2E3 (m)-PR: sc-61749-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.