

UBE2Z siRNA (h): sc-93728

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. UBE2Z (ubiquitin-conjugating enzyme E2 Z), also known as Uba6-specific E2 conjugating enzyme 1 and Ubiquitin carrier protein Z, is a 354 amino acid protein that is the specific substrate for UBE1L2 and may be involved in regulation of apoptosis. Though widely expressed, UBE2Z is found at highest levels in pancreas, testis, placenta and spleen. There are two isoforms of UBE2Z that are produced as a result of alternative splicing events.

REFERENCES

1. Ardley, H.C., Moynihan, T.P., Markham, A.F. and Robinson, P.A. 2000. Promoter analysis of the human ubiquitin-conjugating enzyme gene family UBE2L1-4, including UBE2L3 which encodes Ubch7. *Biochim. Biophys. Acta* 1491: 57-64.
2. Passmore, L.A. and Barford, D. 2004. Getting into position: the catalytic mechanisms of protein ubiquitylation. *Biochem. J.* 379: 513-525.
3. Kuhlbrodt, K., Mouysset, J. and Hoppe, T. 2005. Orchestra for assembly and fate of polyubiquitin chains. *Essays Biochem.* 41: 1-14.
4. Takeuchi, T., Iwahara, S., Saeki, Y., Sasajima, H. and Yokosawa, H. 2005. Link between the ubiquitin conjugation system and the ISG15 conjugation system: ISG15 conjugation to the Ubch6 ubiquitin E2 enzyme. *J. Biochem.* 138: 711-719.
5. Gu, X., Zhao, F., Zheng, M., Fei, X., Chen, X., Huang, S., Xie, Y. and Mao, Y. 2007. Cloning and characterization of a gene encoding the human putative ubiquitin conjugating enzyme E2Z (UBE2Z). *Mol. Biol. Rep.* 34: 183-188.
6. Online Mendelian Inheritance in Man, OMIM[™]. 2007. Johns Hopkins University, Baltimore, MD. MIM Number: 611362. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
7. Napolitano, L.M., Jaffray, E.G., Hay, R.T. and Meroni, G. 2010. Functional interactions between ubiquitin E2 enzymes and TRIM proteins. *Biochem. J.* 434: 309-319.

CHROMOSOMAL LOCATION

Genetic locus: UBE2Z (human) mapping to 17q21.32.

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.

PRODUCT

UBE2Z 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 UBE2Z shRNA Plasmid (h): sc-93728-SH and UBE2Z shRNA (h) Lentiviral Particles: sc-93728-V as alternate gene silencing products.

For independent verification of UBE2Z (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-93728A, sc-93728B and sc-93728C.

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

UBE2Z siRNA (h) is recommended for the inhibition of UBE2Z 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 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 UBE2Z gene expression knockdown using RT-PCR Primer: UBE2Z (h)-PR: sc-93728-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.