

UBA5 siRNA (h): sc-78187

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 ubiquitin-activating enzymes (E1s), the ubiquitin-conjugating enzymes (E2s) and the ubiquitin-protein ligases (E3s). UBA5 (ubiquitin-activating enzyme 5), also known as UBE1DC1 or TH1FP1, is a 404 amino acid protein that belongs to the E1-like ubiquitin-activating enzyme family. Existing as two alternatively spliced isoforms, UBA5 forms a high-energy thioester bond with UFM1 (ubiquitin-fold modifier 1), a protein involved in posttranslational modification. Via formation of a thioester bond, UBA5 activates UFM1 function and may, thus, play a role in the regulation of posttranslational modification events.

REFERENCES

1. Hartley, J.L., et al. 2000. DNA cloning using *in vitro* site-specific recombination. *Genome Res.* 10: 1788-1795.
2. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 610552. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Komatsu, M., et al. 2004. A novel protein-conjugating system for UFM1, a ubiquitin-fold modifier. *EMBO J.* 23: 1977-1986.
4. Dou, T., et al. 2005. Isolation and characterization of ubiquitin-activating enzyme E1-domain containing 1, UBE1DC1. *Mol. Biol. Rep.* 32: 265-271.
5. Sasakawa, H., et al. 2006. Solution structure and dynamics of UFM1, a ubiquitin-fold modifier 1. *Biochem. Biophys. Res. Commun.* 343: 21-26.
6. Mizushima, T., et al. 2007. Crystal structure of Ufc1, the UFM1-conjugating enzyme. *Biochem. Biophys. Res. Commun.* 362: 1079-1084.

CHROMOSOMAL LOCATION

Genetic locus: UBA5 (human) mapping to 3q22.1.

PRODUCT

UBA5 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 UBA5 shRNA Plasmid (h): sc-78187-SH and UBA5 shRNA (h) Lentiviral Particles: sc-78187-V as alternate gene silencing products.

For independent verification of UBA5 (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-78187A, sc-78187B and sc-78187C.

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.

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

UBA5 siRNA (h) is recommended for the inhibition of UBA5 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 UBA5 gene expression knockdown using RT-PCR Primer: UBA5 (h)-PR: sc-78187-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.