SANTA CRUZ BIOTECHNOLOGY, INC.

UBA2 siRNA (h): sc-61740

BACKGROUND

The small ubiquitin-related modifier protein SUMO-1 belongs to the ubiquitin-like protein family, which are synthesized as precursor proteins that undergo processing before conjugation to target proteins. However, SUMO and ubiquitin differ with respect to targeting. Ubiquitination predominantly targets proteins for degradation, whereas sumoylation targets proteins to a variety of cellular processes, including nuclear transport, transcriptional regulation, apoptosis and protein stability. SUMO-1 utilizes homologues of the E1 and E2 enzymes for conjugation to proteins, which include IκBα, MDM2, p53, PML and Ran GAP1. AOS1 is homologous to the N-terminal half of E1 and UBA2 is homologous to the C-terminal half of E1. These proteins form a heterodimer that activates SUMO-1.

REFERENCES

1. Duprez, E., et al. 1999. SUMO-1 modification of the acute promyelocytic leukaemia protein PML: implications for nuclear localisation. J. Cell Sci. 112: 381-393.
2. Gong, L., et al. 1999. Molecular cloning and character the sentrin-activating enzyme complex. FEBS Lett. 448: 185-189.
3. Okuma, T., et al. 1999. *In vitro* SUMO-1 modification requires two enzymatic steps, E1 and E2. Biochem. Biophys. Res. Commun. 254: 693-698.
4. Schwienhorst, I., et al. 2000. SUMO conjugation and deconjugation. Mol. Gen. Genet. 263: 771-786.
5. Saitoh, H., et al. 2000. Functional heterogeneity of small ubiquitin-related protein modifiers SUMO-1 versus SUMO-2/3. J. Biol. Chem. 275: 6252-6258.

CHROMOSOMAL LOCATION

Genetic locus: UBA2 (human) mapping to 19q13.11.

PRODUCT

UBA2 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 UBA2 shRNA Plasmid (h): sc-61740-SH and UBA2 shRNA (h) Lentiviral Particles: sc-61740-V as alternate gene silencing products.

For independent verification of UBA2 (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-61740A, sc-61740B and sc-61740C.

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

UBA2 siRNA (h) is recommended for the inhibition of UBA2 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

UBA2 (B-6): sc-376305 is recommended as a control antibody for monitoring of UBA2 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-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 UBA2 gene expression knockdown using RT-PCR Primer: UBA2 (h)-PR: sc-61740-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Benoit, Y.D., et al. 2021. Targeting SUMOylation dependency in human cancer stem cells through a unique SAE2 motif revealed by chemical genomics. Cell Chem. Biol. 28: 1394-1406.e10.
2. Singhal, J., et al. 2022. Host SUMOylation pathway negatively regulates protective immune responses and promotes *Leishmania donovani* survival. Front. Cell. Infect. Microbiol. 12: 878136.

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.