SANTA CRUZ BIOTECHNOLOGY, INC.

AOS-1 siRNA (m): sc-60175

BACKGROUND

Proteolytic degradation by the ubiquitin (Ub) system is essential for normal cell cycle progression, cellular differentiation and stress responses. Proteins conjugated to Ub are marked for progressive degradation by the 26S Proteasome. AOS-1, also designated SUMO-1-activating enzyme or ubiquitin-like 1-activating enzyme E1A, belongs to the ubiquitin-activating E1 family of proteins and plays an important role in the first step of the UBL1 conjugation pathway. AOS-1, which is a dimeric enzyme, functions as a UBLI E1 ligase, mediating the ATP-dependent activation of UBL1. AOS-1 can bind with UBLE1A and UBLE1B to form a heterodimer which can bind UBL1.

REFERENCES

1. Desterro, J.M., et al. 1998. SUMO-1 modification of IκB-α inhibits NFκB activation. Mol. Cell 2: 233-239.
2. Okuma, T., et al. 1999. *In vitro* SUMO-1 modification requires two enzymatic steps, E1 and E2. Biochem. Biophys. Res. Commun. 254: 693-698.
3. Gong, L., et al. 1999. Molecular cloning and characterization of human AOS-1 and UBA2, components the sentrin-activating enzyme complex. FEBS Lett. 448: 185-189.
4. Desterro, J.M., et al. 1999. Identification of the enzyme required for activation of the small ubiquitin-like protein SUMO-1. J. Biol. Chem. 274: 10618-10624.
5. Engelhardt, O.G., et al. 2001. Interferon-induced antiviral Mx1 GTPase is associated with components of the SUMO-1 system and promyelocytic leukemia protein nuclear bodies. Exp. Cell Res. 271: 286-295.
6. Pichler, A., et al. 2004. The Ran BP-2 SUMO E3 ligase is neither HECT- nor RING-type. Nat. Struct. Mol. Biol.11: 984-991.
7. Lois, L.M., et al. 2005. Structures of the SUMO E1 provide mechanistic insights into SUMO activation and E2 recruitment to E1. EMBO J. 24: 439-451.

CHROMOSOMAL LOCATION

Genetic locus: Sae1 (mouse) mapping to 7 A2.

PRODUCT

AOS-1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see AOS-1 shRNA Plasmid (m): sc-60175-SH and AOS-1 shRNA (m) Lentiviral Particles: sc-60175-V as alternate gene silencing products.

For independent verification of AOS-1 (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-60175A, sc-60175B and sc-60175C.

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 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

AOS-1 siRNA (m) is recommended for the inhibition of AOS-1 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 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

AOS-1 (H-9): sc-398080 is recommended as a control antibody for monitoring of AOS-1 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 AOS-1 gene expression knockdown using RT-PCR Primer: AOS-1 (m)-PR: sc-60175-PR (20 µl). 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.