SANTA CRUZ BIOTECHNOLOGY, INC.

APPBP1 siRNA (h): sc-72523

BACKGROUND

APPBP1 (β-Amyloid precursor protein-binding protein 1), also known as NAE1 (NEDD8-activating enzyme E1 regulatory subunit 1), HPP1 or Ula 1, is a member of the ubiquitin-activating E1 family. In fetal tissues APPBP1 is widely expressed and in adult tissues it is expressed throughout the brain. APPBP1 is a cell membrane associated protein and functions as the regulatory subunit in a heterodimer with UBA3. The APPBP1/UBA3 complex binds to and activates NEDD8, a ubiquitin-like protein involved in signal transduction, cell proliferation and development. This suggests that APPBP1 affects a variety of cellular functions. In addition, APPBP1 is essential for cell cycle progression through the S/M checkpoint. More specifically, it inhibits the entry into S phase and promotes entry into M phase.

REFERENCES

1. Walden, H., et al. 2003. The structure of the APPBP1-UBA3-NEDD8-ATP complex reveals the basis for selective ubiquitin-like protein activation by an E1. Mol. Cell 12: 1427-1437.
2. Bohnsack, R.N. and Haas, A.L. 2003. Conservation in the mechanism of NEDD8 activation by the human APPBP1-UBA3 heterodimer. J. Biol. Chem. 278: 26823-26830.
3. Huang, D.T., et al. 2004. A unique E1-E2 interaction required for optimal conjugation of the ubiquitin-like protein NEDD8. Nat. Struct. Mol. Biol. 11: 927-935.
4. Narasimhan, J., et al. 2005. Crystal structure of the interferon-induced ubiquitin-like protein ISG15. J. Biol. Chem. 280: 27356-27365.
5. Duda, D.M., et al. 2005. Structural analysis of *Escherichia coli* ThiF. J. Mol. Biol. 349: 774-786.
6. Huang, D.T., et al. 2005. Structural basis for recruitment of Ubc12 by an E2 binding domain in NEDD8's E1. Mol. Cell 17: 341-350.
7. Huang, D.T. and Schulman, B.A. 2005. Expression, purification, and characterization of the E1 for human NEDD8, the heterodimeric APPBP1-UBA3 complex. Methods Enzymol. 398: 9-20.
8. Chen, Y., et al. 2007. APP-BP1 inhibits Aβ42 levels by interacting with Presenilin-1. Mol. Neurodegener. 2: 3.

CHROMOSOMAL LOCATION

Genetic locus: APPBP1 (human) mapping to 16q22.1.

PRODUCT

APPBP1 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 APPBP1 shRNA Plasmid (h): sc-72523-SH and APPBP1 shRNA (h) Lentiviral Particles: sc-72523-V as alternate gene silencing products.

For independent verification of APPBP1 (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-72523A, sc-72523B and sc-72523C.

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

APPBP1 siRNA (h) is recommended for the inhibition of APPBP1 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

APPBP1 (C-2): sc-390002 is recommended as a control antibody for monitoring of APPBP1 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 APPBP1 gene expression knockdown using RT-PCR Primer: APPBP1 (h)-PR: sc-72523-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. Xu, D., et al. 2021. Cullin 2-RBX1 E3 ligase and USP2 regulate antithrombin ubiquitination and stability. FASEB J. 35: e21800.

RESEARCH USE

For research use only, not for use in diagnostic procedures.