

AP-2 σ 1 siRNA (m): sc-155865

BACKGROUND

Adapter-related protein complex 2 (AP-2) is one of the two major Clathrin-associated adaptor complexes that links Clathrin to receptors in coated vesicles. It interacts with the cytoplasmic tail of membrane proteins, thereby leading to their selection and concentration. This complex has also been shown to bind polyphosphoinositides. Existing as a heterotetramer, AP-2 is comprised of two large chains, a medium chain and a small chain. Adapter-related protein complex 2 subunit σ -1 (AP-2 σ 1), also known as Clathrin coat assembly protein AP17 or σ 2-Adaptin, is the 142 amino acid protein small chain of the AP-2 complex. Localized to the cytoplasmic side of the cell membrane, two named isoforms of AP-2 σ 1 exist as a result of alternative splicing events.

REFERENCES

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2. Brodsky, F.M., et al. 1991. Clathrin light chains: arrays of protein motifs that regulate coated-vesicle dynamics. *Trends Biochem. Sci.* 16: 208-213.
3. Pley, U. and Parham, P. 1993. Clathrin: its role in receptor-mediated vesicular transport and specialized functions in neurons. *Crit. Rev. Biochem. Mol. Biol.* 28: 431-464.
4. Winterpacht, A., et al. 1996. Human CLAPS2 encoding AP17, a small chain of the Clathrin-associated protein complex: cDNA cloning and chromosomal assignment to 19q13.2→q13.3. *Cytogenet. Cell Genet.* 75: 132-135.
5. Holzmann, K., et al. 1998. A novel spliced transcript of human CLAPS2 encoding a protein alternative to Clathrin adaptor protein AP17. *Gene* 220: 39-44.
6. Online Mendelian Inheritance in Man, OMIM[™]. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 602242. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
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CHROMOSOMAL LOCATION

Genetic locus: Ap2s1 (mouse) mapping to 7 A2.

PRODUCT

AP-2 σ 1 siRNA (m) is a pool of 2 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 AP-2 σ 1 shRNA Plasmid (m): sc-155865-SH and AP-2 σ 1 shRNA (m) Lentiviral Particles: sc-155865-V as alternate gene silencing products.

For independent verification of AP-2 σ 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-155865A and sc-155865B.

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

AP-2 σ 1 siRNA (m) is recommended for the inhibition of AP-2 σ 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 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.

GENE EXPRESSION MONITORING

AP-2 σ 1 (5-RY21): sc-134261 is recommended as a control antibody for monitoring of AP-2 σ 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 AP-2 σ 1 gene expression knockdown using RT-PCR Primer: AP-2 σ 1 (m)-PR: sc-155865-PR (20 μ l). 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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.