SANTA CRUZ BIOTECHNOLOGY, INC.

COPB siRNA (m): sc-41197

BACKGROUND

Membrane and vesicular trafficking in the early secretory pathway are mediated by non-Clathrin COP (coat protein) I-coated vesicles. COPB (β-COP) is a marker protein for pre-Golgi intermediates (vesicular tubular clusters or VTCs). GIV ($G_{\alpha\ i/s}$ interacting protein) co-localizes with COPB and $G_{\alpha\ i3}$ on vesicles found in close proximity to ER exit sites and to *cis* Golgi cisternae. Afadin DIL domain-interacting protein (ADIP) co-localizes with β'-COP (COPP) at the Golgi complex in Madin Darby canine kidney and normal rat kidney cells. Non-Clathrin-coated vesicles mediate membrane traffic through the Golgi complex. COPB is a major component of the coat of non-Clathrin-coated vesicles.

REFERENCES

1. Duden, R., et al. 1991. Involvement of β-COP in membrane traffic through the Golgi complex. Trends Cell Biol. 1: 14-19.
2. Lowe, M., et al. 1995. *In vitro* assembly and dissembly of coatomer. J. Biol. Chem. 270: 31364-31371.
3. Harter, C., et al. 1998. A single binding site for dilysine retrieval motifs and p23 within the γ subunit of coatomer. Proc. Natl. Acad. Sci. USA 95: 11649-11654.
4. Tisdale, E.J., et al. 2003. Atypical protein kinase C plays a critical role in protein transport from pre-Golgi intermediates. J. Biol. Chem. 278: 38015-38021.
5. Cohen, M., et al. 2003. Deubiquitination, a new player in Golgi to endoplasmic reticulum retrograde transport. J. Biol. Chem. 278: 51989-51992.
6. Rybakin, V., et al. 2004. Coronin 7, the mammalian POD-1 homologue, localizes to the Golgi apparatus. FEBS Lett. 573: 161-167.

CHROMOSOMAL LOCATION

Genetic locus: Copb1 (mouse) mapping to 7 F1.

PRODUCT

COPB 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 COPB shRNA Plasmid (m): sc-41197-SH and COPB shRNA (m) Lentiviral Particles: sc-41197-V as alternate gene silencing products.

For independent verification of COPB (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-41197A, sc-41197B and sc-41197C.

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

COPB siRNA (m) is recommended for the inhibition of COPB 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

COPB (D-10): sc-393615 is recommended as a control antibody for monitoring of COPB 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 COPB gene expression knockdown using RT-PCR Primer: COPB (m)-PR: sc-41197-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. Casey, C.A., et al. 2016. Study of ethanol-induced Golgi disorganization reveals the potential mechanism of alcohol-impaired N-glycosylation. Alcohol. Clin. Exp. Res. 40: 2573-2590.

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.