SANTA CRUZ BIOTECHNOLOGY, INC.

COPG siRNA (h): sc-37254

BACKGROUND

Membrane and vesicular trafficking in the early secretory pathway are mediated by non-clathrin COP (coat protein) I-coated vesicles. COPI-coated vesicles mediate retrograde transport from the Golgi back to the ER and intra-Golgi transport. The cytosolic precursor of the COPI coat, the heptameric coatomer complex, is composed of two subcomplexes. The first consists of the COPB, COPG, COPD and COPZ subunits (also known as β-, γ-, δ- and ζ-COP, respectively), which are distantly homologous to AP Clathrin adaptor subunits. The second consists of the COPA, β'-COP and COPE subunits (also known as α-COP, COPP and ε-COP, respectively). The COPG (γ-COP) subunit of the coatomer is believed to mediate the binding to the cytoplasmic dilysine motifs of membrane proteins. COPG has the highest level of expression in mouse testis and is expressed in a parent-of-origin-specific manner in mammals.

REFERENCES

1. Stenbeck, G., et al. 1992. γ-COP, a COAT subunit of non-clathrin-coated vesicles with homology to Sec21p. FEBS Lett. 314: 195-198.
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. Hahn, Y., et al. 2000. Duplication of genes encoding non-clathrin coat protein γ-COP in vertebrate, insect and plant evolution. FEBS Lett. 482: 31-36.
5. Futatsumori, M., et al. 2000. Identification and characterization of novel isoforms of COP I subunits. J. Biochem. 128: 793-801.
6. Bermak, J.C., et al. 2002. Interaction of γ-COP with a transport motif in the D1 receptor C-terminus. Eur. J. Cell Biol. 81: 77-85.
7. Watson, P.J., et al. 2004. γ-COP appendage domain—structure and function. Traffic 5: 79-88.
8. SWISS-PROT/TrEMBL (P48444). World Wide Web URL: http://harvester.embl.de/harvester/P484/P48444.htm

CHROMOSOMAL LOCATION

Genetic locus: COPG1 (human) mapping to 3q21.3.

PRODUCT

COPG 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 COPG shRNA Plasmid (h): sc-37254-SH and COPG shRNA (h) Lentiviral Particles: sc-37254-V as alternate gene silencing products.

For independent verification of COPG (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-37254A, sc-37254B and sc-37254C.

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

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

COPG (A-10): sc-393977 is recommended as a control antibody for monitoring of COPG 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 COPG gene expression knockdown using RT-PCR Primer: COPG (h)-PR: sc-37254-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.

PROTOCOLS

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