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COPA siRNA (h): sc-43696

BACKGROUND

COPA (α -coat protein) is processed to produce Xenin. Xenin stimulates exocrine pancreatic secretion to affect small and large intestinal motility, and inhibits pentagastrin-stimulated secretion of acid. In the gut, Xenin interacts with the neurotensin receptor. 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).

REFERENCES

1. Quek, H.H., et al. 1997. Molecular and cellular studies of the human homolog of the 160-kD α -subunit of the coatomer protein complex. *DNA Cell Biol.* 16: 275-280
2. Chow, V.T., et al. 1997. α coat protein COPA (HEP-COP): presence of an Alu repeat in cDNA and identity of the amino terminus to xenin. *Ann. Hum. Genet.* 61: 369-373.
3. Feurle, G.E., et al. 1998. Xenin—a review. *Peptides* 19: 609-615.
4. Schroder-Kohne, S., et al. 1998. α -COP can discriminate between distinct, functional dilysine signals *in vitro* and regulates access into retrograde transport. *J. Cell Sci.* 111: 3459-3470.
5. Chaudhary, A., et al. 1998. Specific interaction of Golgi coatomer protein α -COP with phosphatidylinositol 3,4,5-trisphosphate. *J. Biol. Chem.* 273: 8344-8350.

CHROMOSOMAL LOCATION

Genetic locus: COPA (human) mapping to 1q23.2.

PRODUCT

COPA 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see COPA shRNA Plasmid (h): sc-43696-SH and COPA shRNA (h) Lentiviral Particles: sc-43696-V as alternate gene silencing products.

For independent verification of COPA (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-43696A, sc-43696B and sc-43696C.

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.

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

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

COPA (H-3): sc-398099 is recommended as a control antibody for monitoring of COPA 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 COPA gene expression knockdown using RT-PCR Primer: COPA (h)-PR: sc-43696-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.