SANTA CRUZ BIOTECHNOLOGY, INC.

CPA2 siRNA (h): sc-89757

BACKGROUND

Members of the M14 metallocarboxypeptidase protein family serve many diverse functions and are divided into three subfamilies based on structure, function and amino acid sequence similarity. As a member of the A/B subfamily, CPA2 (Carboxypeptidase A2) is a 417 amino acid zinc-binding secreted protein that contains a characteristic propetide at the amino-terminus, which is cleaved off upon enzyme activation. CPA2 is similar to CPA1, a pancreatic exopeptidase that catalyzes the release of C-terminal amino acids from a variety of proteins, thereby playing a key role in protein digestion and degradation. CPA1 and CPA2 differ in their substrate specificities with CPA2 preferring bulkier C-terminal residues. Expression of CPA2 has been detected in pancreas, brain, lung and testis.

REFERENCES

1. Gardell, S.J., et al. 1988. A novel rat carboxypeptidase, CPA2: characterization, molecular cloning, and evolutionary implications on substrate specificity in the carboxypeptidase gene family. J. Biol. Chem. 263: 17828-17836.
2. Clauser, E., et al. 1988. Structural characterization of the rat carboxypeptidase A1 and B genes. Comparative analysis of the rat carboxypeptidase gene family. J. Biol. Chem. 263: 17837-17845.
3. Moulard, M., et al. 1990. Further studies on the human pancreatic binary complexes involving procarboxypeptidase A. FEBS Lett. 261: 179-183.
4. Faming, Z., et al. 1991. Structural evolution of an enzyme specificity. The structure of rat carboxypeptidase A2 at 1.9-A resolution. J. Biol. Chem. 266: 24606-24612.
5. Linder, D., et al. 1993. Separation of human pancreatic carboxypeptidase A isoenzymes by high performance liquid chromatography. Biomed. Chromatogr. 7: 143-145.
6. Normant, E., et al. 1995. Carboxypeptidase A isoforms produced by distinct genes or alternative splicing in brain and other extrapancreatic tissues. J. Biol. Chem. 270: 20543-20549.
7. Laethem, R.M., et al. 1996. Expression and characterization of human pancreatic preprocarboxypeptidase A1 and preprocarboxypeptidase A2. Arch. Biochem. Biophys. 332: 8-18.

CHROMOSOMAL LOCATION

Genetic locus: CPA2 (human) mapping to 7q32.2.

PRODUCT

CPA2 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 CPA2 shRNA Plasmid (h): sc-89757-SH and CPA2 shRNA (h) Lentiviral Particles: sc-89757-V as alternate gene silencing products.

For independent verification of CPA2 (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-89757A, sc-89757B and sc-89757C.

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

CPA2 siRNA (h) is recommended for the inhibition of CPA2 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

CPA2 (B-5): sc-393547 is recommended as a control antibody for monitoring of CPA2 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 CPA2 gene expression knockdown using RT-PCR Primer: CPA2 (h)-PR: sc-89757-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.