

CA III siRNA (h): sc-60309

BACKGROUND

Carbonic anhydrases (CAs) are members of a large family of zinc metalloenzymes that catalyze the reversible hydration of carbon dioxide. CAs are involved in a variety of biological processes, including respiration, calcification, acid-base balance, bone resorption, and the formation of aqueous humor, cerebrospinal fluid, saliva, and gastric juice. They show extensive diversity in distribution and in their subcellular localization. CA III (carbonic anhydrase III), also known as Car3 or CA3, is a 260 amino acid cytoplasmic protein that is specifically expressed in muscle. Belonging to the α -carbonic anhydrase family, CA III is activated by proton donors such as imidazole and dipeptide histidylhistidine, and is inhibited by coumarins and sulfonamide derivatives such as acetazolamide.

REFERENCES

1. Heath, R., et al. 1985. Evaluation of carrier detection of Duchenne muscular dystrophy using carbonic anhydrase III and creatine kinase. *Am. J. Med. Genet.* 21: 291-296.
2. Edwards, Y.H., et al. 1988. The gene for human muscle specific carbonic anhydrase (CA III) is assigned to chromosome 8. *Ann. Hum. Genet.* 50: 41-47.
3. Beechey, C., et al. 1990. Mapping of mouse carbonic anhydrase-3, Car3: another locus in the homologous region of mouse chromosome 3 and human chromosome 8. *Genomics* 6: 692-696.
4. Igarashi, S., et al. 1992. Comparison of the distribution of carbonic anhydrase isozymes (CA I, CA II, CA III) in the rat gastrointestinal tract. *J. Vet. Med. Sci.* 54: 535-539.
5. Mahieu, I., et al. 1995. Localisation of CA II, CA III and CA IV in an umbilical vein endothelial cell line (EA-hy926). *Biochem. Soc. Trans.* 23: 308S.
6. Rusconi, S., et al. 2004. Carbonic anhydrase inhibitors. Interaction of isozymes I, II, IV, V, and IX with phosphates, carbamoyl phosphate, and the phosphonate antiviral drug foscarnet. *Bioorg. Med. Chem. Lett.* 14: 5763-5767.
7. Innocenti, A., et al. 2005. Carbonic anhydrase inhibitors. Inhibition of isozymes I, II, IV, V, and IX with anions isosteric and isoelectronic with sulfate, nitrate, and carbonate. *Bioorg. Med. Chem. Lett.* 15: 567-571.

CHROMOSOMAL LOCATION

Genetic locus: CA3 (human) mapping to 8q21.2.

PRODUCT

CA III 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 CA III shRNA Plasmid (h): sc-60309-SH and CA III shRNA (h) Lentiviral Particles: sc-60309-V as alternate gene silencing products.

For independent verification of CA III (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-60309A, sc-60309B and sc-60309C.

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

CA III siRNA (h) is recommended for the inhibition of CA III 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

CA III (F-10): sc-373729 is recommended as a control antibody for monitoring of CA III 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 CA III gene expression knockdown using RT-PCR Primer: CA III (h)-PR: sc-60309-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.