

CA IV siRNA (h): sc-29867

BACKGROUND

Carbonic anhydrase IV (CA IV) is glycosylphosphatidylinositol-anchored to the outer surface of the plasma membrane where it catalyzes hydration-dehydration of $\text{CO}_2/\text{HCO}_3^-$. CA IV is present on the plasma face of microcapillaries and in the choriocapillaris of the human eye. CA IV facilitates renal acidification in the kidney and is responsible for the regulation of interstitial pH (pH_i) transients in brain. Impairment in targeting leads to disruption of HCO_3^- secretion and associates with malfunction in cystic fibrosis cells. Carbonic anhydrases are zinc metalloenzymes that catalyze the reversible hydration of carbon dioxide. They participate in respiration, calcification, acid-base balance, bone resorption, and the formation of aqueous humor, cerebrospinal fluid, saliva, and gastric acid. Carbonic anhydrases show extensive diversity in tissue distribution and in their subcellular localization.

REFERENCES

1. Tong, C.K., et al. 2000. Interstitial carbonic anhydrase (CA) activity in brain is attributable to membrane-bound CA type IV. *J. Neurosci.* 20: 8247-8253.
2. Schwartz, G.J., et al. 2002. Carbonic anhydrase XII mRNA encodes a hydratase that is differentially expressed along the rabbit nephron. *Am. J. Physiol. Renal Physiol.* 284: F399-F410.
3. Fanjul, M., et al. 2002. Targeting of carbonic anhydrase IV to plasma membranes is altered in cultured human pancreatic duct cells expressing a mutated (δF508) CFTR. *Eur. J. Cell Biol.* 81: 437-447.
4. Sterling, D., et al. 2002. The extracellular component of a transport metabolon. Extracellular loop 4 of the human AE1 $\text{Cl}^-/\text{HCO}_3^-$ exchanger binds carbonic anhydrase IV. *J. Biol. Chem.* 277: 25239-25246.
5. Alvarez, B.V., et al. 2003. Direct extracellular interaction between carbonic anhydrase IV and the human NBC1 sodium/bicarbonate co-transporter. *Biochemistry* 42: 12321-12329.

CHROMOSOMAL LOCATION

Genetic locus: CA4 (human) mapping to 17q23.1.

PRODUCT

CA IV 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 CA IV shRNA Plasmid (h): sc-29867-SH and CA IV shRNA (h) Lentiviral Particles: sc-29867-V as alternate gene silencing products.

For independent verification of CA IV (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-29867A, sc-29867B and sc-29867C.

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 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

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

CA IV (H-5): sc-74446 is recommended as a control antibody for monitoring of CA IV 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 IV gene expression knockdown using RT-PCR Primer: CA IV (h)-PR: sc-29867-PR (20 μl). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

RESEARCH USE

For research use only, not for use in diagnostic procedures.