Store at -20°C

SignalSilence® PKA C-α siRNA I

#6406

10 µM in 300 µl (3 nmol)

Support: +1-978-867-2388 (U.S.)
www.cellsignal.com/support

Orders: 877-616-2355 (U.S.)
orders@cellsignal.com

Entrez-Gene ID #5566
UniProt ID #P17612

rev. 02/10/16

For Research Use Only. Not For Use In Diagnostic Procedures.

Species Cross-Reactivity: H

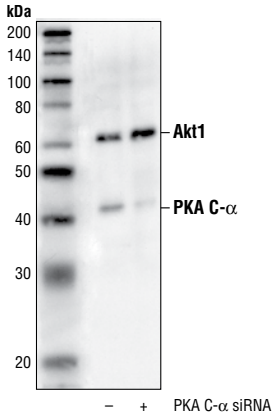
Description: SignalSilence® PKA C-α siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit PKA C-α expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules into the cell. All SignalSilence® siRNA products from CST are rigorously tested in-house and have been shown to reduce target protein expression by western analysis.

Background: The second messenger cyclic AMP (cAMP) activates cAMP-dependent protein kinase (PKA or cAPK) in mammalian cells and controls many cellular mechanisms such as gene transcription, ion transport, and protein phosphorylation (1). Inactive PKA is a heterotetramer composed of a regulatory subunit (R) dimer and a catalytic subunit (C) dimer. In this inactive state, the pseudosubstrate sequences on the R subunits block the active sites on the C subunits. Three C subunit isoforms (C-α, C-β, and C-γ) and two families of regulatory subunits (RI and RII) with distinct cAMP binding properties have been identified. The two R families exist in two isoforms, α and β (RI-α, RI-β, RII-α, and RII-β). Upon binding of cAMP to the R subunits, the autoinhibitory contact is eased and active monomeric C subunits are released. PKA shares substrate specificity with Akt (PKB) and PKC, which are characterized by an arginine at position -3 relative to the phosphorylated serine or threonine residue (2). Substrates that present this consensus sequence and have been shown to be phosphorylated by PKA are Bad (Ser155), CREB (Ser133), and GSK-3 (GSK-3α Ser21 and GSK-3β Ser9) (3-5). In addition, combined knock-down of PKA C-α and -β blocks cAMP-mediated phosphorylation of Raf (Ser43 and Ser259) (6). Autophosphorylation and phosphorylation by PDK-1 are two known mechanisms responsible for phosphorylation of the C subunit at Thr197 (7).

Small interfering RNA (siRNA) has been used to specifically silence PKA C-α in NIH/3T3 cells (6).

Directions for Use: CST recommends transfection with 100 nM SignalSilence® PKA C-α siRNA I 48 hours prior to cell lysis. For transfection procedure, follow protocol provided by the transfection reagent manufacturer. Please feel free to contact CST with any questions on use.

Limited Use Label License, RNA interference: This product is licensed under European Patent 1144623 and foreign equivalents from Ribopharma AG, Kulmbach, Germany and is provided only for use in non-commercial research specifically excluding use (a) in



Western blot analysis of extracts from HeLa cells, transfected with non-targeted siRNA (-) or SignalSilence® PKA C-α siRNA I (+), using PKA C-α Antibody #4782 and Akt1 (2H10) Mouse mAb #2967. The PKA C-α antibody confirms silencing of PKA C-α expression, while the Akt1 antibody is used to control for loading and specificity of PKA C-α siRNA.t

Storage: PKA C-α siRNA I is supplied in RNAse-free water. *Aliquot and store at -20°C.*

For product specific protocols and a complete listing of recommended companion products please see the product web page at www.cellsignal.com

Background References:

(1) Montminy, M. (1997) *Annu. Rev. Biochem.* 66, 807-822.

(2) Dell'Acqua, M.L. and Scott, J.D. (1997) *J. Biol. Chem.* 272, 12881-12884.

(3) Tan, Y. et al. (2000) *J. Biol. Chem.* 275, 25865-25869.

(4) Gonzalez, G.A. and Montminy, M.R. (1989) *Cell* 59, 675-680.

(5) Fang, X. et al. (2000) *Proc. Natl. Acad. Sci. USA* 97, 11960-11965.

(6) Dumaz, N. and Marais, R. (2003) *J. Biol. Chem.* 278, 29819 -29823.

(7) Moore, M.J. et al. (2002) *J. Biol. Chem.* 277, 47878-47884.

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Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity:** **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—Horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.