#13553 Store at -20°C

SignalSilence® Sara siRNA II

Cell Signaling TECHNOLOGY®

Orders ■ 877-616-CELL (2355)
orders@cellsignal.com
Support ■ 877-678-TECH (8324)
info@cellsignal.com
Web ■ www.cellsignal.com

New 12/13

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

Entrez-Gene ID #9372
UniProt Acc. #O95405

Species Cross-Reactivity: H

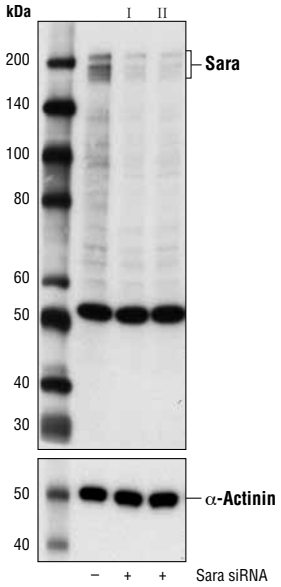
Description: SignalSilence® Sara siRNA II from Cell Signaling Technology (CST) allows the researcher to specifically inhibit Sara expression using RNA interference, a method whereby gene expression can be selectively silenced through the delivery of double stranded RNA molecules into the cell. All SignalSilence®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

Background: The Smad anchor for receptor activation (SARA, ZFYVE9) protein is an FYVE domain-containing protein originally identified as a regulator of TGF-β signaling (1). FYVE domains are zinc finger-like domains that bind to phosphatidylinositol 3-phosphate and are responsible for endosomal trafficking (2). While the role of Sara in TGF-β signaling has been questioned (3,4), early research studies demonstrate that Sara enhances TGF-β signaling by binding and recruiting non-activated Smad2 and Smad3 to the TGF-β receptor complex (1). Upon Smad2 activation, Sara dissociates from the complex while phosphorylated Smad2/3 translocates to the nucleus to bind to the common Smad, Smad4. Sara can also function as an anchor for the protein phosphatase 1 (PP1c) catalytic subunit, which is involved in the Smad7-mediated dephosphorylation of TGF-β type I receptor (5,6). Additional research studies show that expression of Sara plays a critical role in maintenance of the epithelial cell phenotype and that expression is regulated during the epithelial-to-mesenchymal transition (EMT) and fibrosis (7,8).

Directions for Use: CST recommends transfection with 100 nM SignalSilence® Sara siRNA II 48 to 72 hours prior to cell lysis. For transfection procedure, follow the protocol provided by the transfection reagent manufacturer. Please feel free to contact CST with any questions on use.

Each vial contains the equivalent of 100 transfections, which corresponds to a final siRNA concentration of 100 nM per transfection in a 24-well plate with a total volume of 300 µl per well.

Quality Control: Oligonucleotide synthesis is monitored base by base through trityl analysis to ensure appropriate coupling efficiency. The oligo is subsequently purified by affinity-solid phase extraction. The annealed RNA duplex is further analyzed by mass spectrometry to verify the exact composition of the duplex. Each lot is compared to the previous lot by mass spectrometry to ensure maximum lot-to-lot consistency.



Western blot analysis of extracts from SH-SY-5Y cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® Sara siRNA I #13542 (+) or SignalSilence® Sara siRNA II (+), using Sara (D5X4F) Rabbit mAb #13285 (upper) or α-Actinin (D6F6) XP® Rabbit mAb #6487 (lower). The Sara (D5X4F) Rabbit mAb confirms silencing of Sara expression, while the α-Actinin (D6F6) XP® Rabbit mAb is used as a loading control.

Storage: Sara siRNA II is supplied in RNAse-free water. *Aliquot and store at -20°C.*

For product specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

Background References:

(1) Tsukazaki, T. et al. (1998) *Cell* 95, 779-91.
(2) Itoh, F. et al. (2002) *Genes Cells* 7, 321-31.
(3) Bakkebø, M. et al. (2012) *FEBS Lett* 586, 3367-72.
(4) Goto, D. et al. (2001) *Biochem Biophys Res Commun* 281, 1100-5.
(5) Bennett, D. and Alphey, L. (2002) *Nat Genet* 31, 419-23.
(6) Shi, W. et al. (2004) *J Cell Biol* 164, 291-300.
(7) Runyan, C.E. et al. (2009) *J Biol Chem* 284, 25181-9.
(8) Zhao, B.M. and Hoffmann, F.M. (2006) *Mol Biol Cell* 17, 3819-31.

Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **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.