

SignalSilence® EWS siRNA II

✓ 10 µM in 300 µl (3 nmol)

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

New 11/12

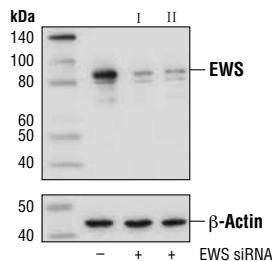
For Research Use Only. Not For Use In Diagnostic Procedures.

Species Cross-Reactivity: H, (M, R, Mk)

Description: SignalSilence® EWS siRNA II from Cell Signaling Technology (CST) allows the researcher to specifically inhibit EWS 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 Ewing sarcoma (EWS) protein is a member of the multifunctional FET (FUS, EWS, and TAF15) family of proteins (1,2). These proteins are RNA and DNA binding proteins that are thought to be important for both transcriptional regulation and RNA processing. EWS can be found as part of a fusion protein with various E-twenty six (ETS) family transcription factors, most commonly Fli-1, in the Ewing sarcoma family of tumors (1-4). The amino terminus of the EWS protein, containing the transcriptional activation domain, is fused to the DNA binding domain of the ETS transcription factor, causing aberrant expression of target genes (1-5). EWS interacts with the transcription initiation complex via TFIID and RNA polymerase II subunits, as well as transcriptional regulators, such as Brn3A and CBP/p300, which suggests a role for EWS in transcriptional regulation (1,6-9). EWS also interacts with multiple components of the splicing machinery, implicating a role for EWS in RNA processing (1,10-12). EWS regulates the expression of cyclin D1, which controls G1-S phase transition during the cell cycle, at the level of transcriptional activation and mRNA splicing. The EWS-Fli-1 fusion protein has been shown to promote the expression of the cyclin D1b splice variant in Ewing sarcoma cells (13). In addition, EWS regulates the DNA damage-induced alternative splicing of genes involved in DNA repair and stress response and is required for cell viability upon DNA damage (14). Consistent with these results, EWS knockout mice display hypersensitivity to ionizing radiation and premature cellular senescence, suggesting a role for EWS in homologous recombination and maintenance of genomic stability (15).

Specificity/Sensitivity: SignalSilence® EWS siRNA II inhibits human, mouse, rat, and monkey EWS expression.



Western blot analysis of extracts from HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-), SignalSilence® EWS siRNA I #12175 (+), or SignalSilence® EWS siRNA II (+), using EWS Antibody #11910 (upper) or β-Actin (D6A8) Rabbit mAb #8457 (lower). The EWS Antibody confirms silencing of EWS expression, while the β-Actin (D6A8) Rabbit mAb is used as a loading control.

Directions for Use: CST recommends transfection with 100 nM SignalSilence® EWS siRNA II 48 to 72 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.

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.

Entrez-Gene ID #2130
Swiss-Prot Acc. #Q01844

Storage: EWS siRNA II is supplied in RNase-free water. Aliquot and store at -20°C.

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

Background References:

- (1) Law, W.J. et al. (2006) *Brief Funct Genomic Proteomic* 5, 8-14.
- (2) Kovar, H. (2011) *Sarcoma* 2011, 837474.
- (3) Delattre, O. et al. (1992) *Nature* 359, 162-5.
- (4) May, W.A. et al. (1993) *Mol Cell Biol* 13, 7393-8.
- (5) Sorensen, P.H. et al. (1994) *Nat Genet* 6, 146-51.
- (6) Bertolotti, A. et al. (1996) *EMBO J* 15, 5022-31.
- (7) Bertolotti, A. et al. (1998) *Mol Cell Biol* 18, 1489-97.
- (8) Araya, N. et al. (2003) *J Biol Chem* 278, 5427-32.
- (9) Thomas, G.R. and Latchman, D.S. *Cancer Biol Ther* 1, 428-32.
- (10) Chansky, H.A. et al. (2001) *Cancer Res* 61, 3586-90.
- (11) Yang, L. et al. (2000) *J Biol Chem* 275, 37612-8.
- (12) Knoop, L.L. and Baker, S.J. (2001) *J Biol Chem* 276, 22317-22.
- (13) Sanchez, G. et al. (2008) *Proc Natl Acad Sci U S A* 105, 6004-9.
- (14) Paronetto, M.P. et al. (2011) *Mol Cell* 43, 353-68.
- (15) Li, H. et al. (2007) *J Clin Invest* 117, 1314-23.