

SignalSilence® USP9X siRNA I



✓ 10 µM 300 µl
(100 transfections)

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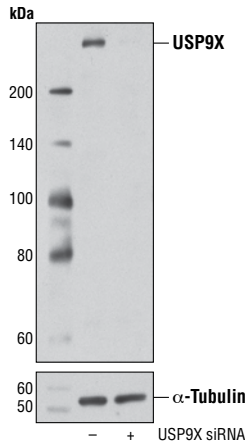
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Species Cross-Reactivity: H

Description: SignalSilence® USP9X siRNA I from Cell Signaling Technology (CST) allows the researcher to specifically inhibit USP9X 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: Protein ubiquitination and deubiquitination are reversible processes catalyzed by ubiquitinating enzymes (UBEs) and deubiquitinating enzymes (DUBs) respectively (1,2). DUBs are categorized into five subfamilies—USP, UCH, OTU, MJD, and JAMM. Ubiquitin-specific protease 9, X-linked (USP9X) possesses a well-conserved catalytic domain with cysteine peptidase activity, which allows for cleavage of ubiquitin and polyubiquitin conjugates. USP9X is the mammalian homolog of the *Drosophila fat-facets (faf)* gene, which is essential for normal eye development and viability of the early fly embryo (3,4). While USP9X expression is also critical for normal mammalian development (5-7), many of its substrates are only beginning to be elucidated. There is mounting evidence that USP9X functions in the formation of epithelial cell-cell contacts through deubiquitination-dependent stabilization of molecules involved in maintaining the integrity of both adherens and tight junctions. Indeed, USP9X has been found to associate with AF-6, the β-catenin-E-cadherin complex, and EFA6 (8-11). Research studies have also demonstrated that USP9X is an integral component of the TGF-β/BMP signaling cascade by opposing TRIM33-mediated monoubiquitination of SMAD4 (12). USP9X is overexpressed in a variety of human cancers and contributes to enhanced cell survival, in part, through its ability to deubiquitinate and stabilize the Mcl-1 oncoprotein (13). There is some evidence, however, that suggests the role of USP9X in tumorigenesis is context dependent. Research studies have implicated USP9X in a tumor suppressor role during the early stages of pancreatic ductal adenocarcinoma (PDAC) and in an oncogenic role during advanced stages of PDAC (14,15).

Directions for Use: CST recommends transfection with 100 nM USP9X siRNA I 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.



Western blot analysis of extracts from HeLa cells, transfected with 100 nM SignalSilence® Control siRNA (Unconjugated) #6568 (-) or SignalSilence® USP9X siRNA I (+), using USP9X Antibody #5751 (upper) or α-Tubulin (11H10) Rabbit mAb #2125 (lower). The USP9X Antibody confirms silencing of USP9X expression, while the α-Tubulin (11H10) Rabbit mAb is used as a loading control.

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 #8239
Swiss-Prot Acc. #Q93008

Storage: USP9X siRNA I is supplied in RNase-free water. Aliquot and store at -20°C.

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

Background References:

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- (15) Cox, J.L. et al. (2014) *Cancer Biol Ther* 15, 1042-52.