

# USP22 siRNA (h): sc-63195

## BACKGROUND

The ubiquitin (Ub) pathway involves three sequential enzymatic steps that facilitate the conjugation of Ub and Ub-like molecules to specific protein substrates. Through the use of a wide range of enzymes that can add or remove ubiquitin, the Ub pathway controls many intracellular processes such as signal transduction, transcriptional activation and cell cycle progression. USP22 (ubiquitin specific peptidase 22), also known as USP3L, is a 525 amino acid protein that contains one UBP-type zinc-finger and functions to catalyze the conversion of a ubiquitin C-terminal thioester to free ubiquitin and thiol, a reaction that may influence several cellular processes. Via its catalytic activity, USP22 is thought to play an important role in cell cycle progression and may also serve as a cancer stem cell marker. The gene encoding USP22 maps to human chromosome 17p11.2, which comprises over 2.5% of the human genome and encodes over 1,200 genes.

## REFERENCES

1. Kikuno, R., et al. 1999. Prediction of the coding sequences of unidentified human genes. XIV. The complete sequences of 100 new cDNA clones from brain which code for large proteins *in vitro*. *DNA Res.* 6: 197-205.
2. Lee, H.J., et al. 2006. The expression patterns of deubiquitinating enzymes, USP22 and Usp22. *Gene Expr. Patterns* 6: 277-284.
3. Zhang, X.Y., et al. 2008. USP22, an hSAGA subunit and potential cancer stem cell marker, reverses the polycomb-catalyzed ubiquitylation of Histone H2A. *Cell Cycle* 7: 1522-1524.
4. Zhao, Y., et al. 2008. A TFTC/STAGA module mediates Histone H2A and H2B deubiquitination, coactivates nuclear receptors and counteracts heter-ochromatin silencing. *Mol. Cell* 29: 92-101.

## CHROMOSOMAL LOCATION

Genetic locus: USP22 (human) mapping to 17p11.2.

## PRODUCT

USP22 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see USP22 shRNA Plasmid (h): sc-63195-SH and USP22 shRNA (h) Lentiviral Particles: sc-63195-V as alternate gene silencing products.

For independent verification of USP22 (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-63195A, sc-63195B and sc-63195C.

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

USP22 siRNA (h) is recommended for the inhibition of USP22 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  $\mu$ M in 66  $\mu$ l. 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

USP22 (C-3): sc-390585 is recommended as a control antibody for monitoring of USP22 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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 $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 USP22 gene expression knockdown using RT-PCR Primer: USP22 (h)-PR: sc-63195-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Kim, D., et al. 2017. Deubiquitinating enzyme USP22 positively regulates c-Myc stability and tumorigenic activity in mammalian and breast cancer cells. *J. Cell. Physiol.* 232: 3664-3676.
2. Gong, Z., et al. 2018. Identification of potential target genes of USP22 via ChIP-seq and RNA-seq analysis in HeLa cells. *Genet. Mol. Biol.* 41: 488-495.

## RESEARCH USE

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

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.