

USP52 siRNA (m): sc-76874

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. USP52 (ubiquitin specific peptidase 52), also known as PAN2 (PAB-dependent poly(A)-specific ribonuclease subunit 2), is a 1,202 amino acid cytoplasmic and nuclear protein belonging to the peptidase C19 family. Containing one exonuclease domain, USP52 is involved in cytoplasmic mRNA decay. USP52 is a component of the Pan nuclease complex, which shortens poly(A) tails of RNA when the poly(A) stretch is bound by the polyadenylate-binding protein.

REFERENCES

1. Boeck, R., Tarun, S., Rieger, M., Deardorff, J.A., Müller-Auer and S., Sachs, A.B. 1996. The yeast Pan2 protein is required for poly(A)-binding protein-stimulated poly(A)-nuclease activity. *J. Biol. Chem.* 271: 432-438.
2. D'Andrea, A., Pellman, D. 1998. Deubiquitinating enzymes: a new class of biological regulators. *Crit. Rev. Biochem. Mol. Biol.* 33: 337-352.
3. Chung, C.H., Baek, S.H. 1999. Deubiquitinating enzymes: their diversity and emerging roles. *Biochem. Biophys. Res. Commun.* 266: 633-640.
4. Hammet, A., Pike and B.L., Heierhorst, J. 2002. Posttranscriptional regulation of the RAD5 DNA repair gene by the Dun1 kinase and the Pan2-Pan3 poly(A)-nuclease complex contributes to survival of replication blocks. *J. Biol. Chem.* 277: 22469-22474.
5. Edmonds, M. 2002. A history of poly A sequences: from formation to factors to function. *Prog. Nucleic Acid Res. Mol. Biol.* 71: 285-389.
6. Yamashita, A., Chang, T.C., Yamashita, Y., Zhu, W., Zhong, Z., Chen and C.Y., Shyu, A.B. 2005. Concerted action of poly(A) nucleases and decapping enzyme in mammalian mRNA turnover. *Nat. Struct. Mol. Biol.* 12: 1054-1063.

CHROMOSOMAL LOCATION

Genetic locus: Pan2 (mouse) mapping to 10 D3.

PRODUCT

USP52 siRNA (m) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see USP52 shRNA Plasmid (m): sc-76874-SH and USP52 shRNA (m) Lentiviral Particles: sc-76874-V as alternate gene silencing products.

For independent verification of USP52 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-76874A, sc-76874B and sc-76874C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

APPLICATIONS

USP52 siRNA (m) is recommended for the inhibition of USP52 expression in mouse 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 μ M in 66 μ 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

USP52 (6A7): sc-517176 is recommended as a control antibody for monitoring of USP52 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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 USP52 gene expression knockdown using RT-PCR Primer: USP52 (m)-PR: sc-76874-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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