

hnRNP K siRNA (m): sc-38283

BACKGROUND

Heterogeneous nuclear ribonucleoproteins (hnRNPs) constitute a set of polypeptides that contribute to mRNA transcription and pre-mRNA processing as well as mature mRNA transport to the cytoplasm and translation. They also bind heterogeneous nuclear RNA (hnRNA), which are the transcripts produced by RNA Polymerase II. There are approximately 20 known hnRNP proteins and their complexes are the major constituents of the spliceosome. The majority of hnRNP protein are localized to the nucleus, however some shuttle between the nucleus and the cytoplasm, such as hnRNP K. hnRNP K recruits a variety of molecular partners through two K homologous (KH) domains, which are required for protein-protein interactions. hnRNP K also contains several potential phosphorylation sites, including Ser 302, the major site of PKC δ phosphorylation, which are thought to regulate various cellular functions, including sequence-specific DNA binding, transcription, RNA binding and nucleocytoplasmic shuttling.

REFERENCES

1. Siomi, H., et al. 1993. The pre-mRNA binding K protein contains a novel evolutionarily conserved motif. *Nucleic Acids Res.* 21: 1193-1198.
2. Badolato, J., et al. 1995. Identification and characterisation of a novel human RNA-binding protein. *Gene* 166: 323-337.
3. Siomi, H., et al. 1995. A nuclear localization domain in the hnRNP A1 protein. *J. Cell Biol.* 129: 551-560.
4. Schullery, D.S., et al. 1999. Regulated interaction of protein kinase C δ with the heterogeneous nuclear ribonucleoprotein K protein. *J. Biol. Chem.* 274: 15101-15109.
5. Kim, J.H., et al. 2000. Protein-protein interaction among hnRNPs shuttling between nucleus and cytoplasm. *J. Mol. Biol.* 298: 395-405.
6. Melcak, I., et al. 2000. Nuclear pre-mRNA compartmentalization: trafficking of released transcripts to splicing factor reservoirs. *Mol. Biol. Cell* 11: 497-510.
7. Mandal, M., et al. 2001. Growth factors regulate heterogeneous nuclear ribonucleoprotein K expression and function. *J. Biol. Chem.* 276: 9699-9704.

CHROMOSOMAL LOCATION

Genetic locus: Hnmpk (mouse) mapping to 13 B1.

PRODUCT

hnRNP K 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 hnRNP K shRNA Plasmid (m): sc-38283-SH and hnRNP K shRNA (m) Lentiviral Particles: sc-38283-V as alternate gene silencing products.

For independent verification of hnRNP K (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-38283A, sc-38283B and sc-38283C.

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

hnRNP K siRNA (m) is recommended for the inhibition of hnRNP K 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

p-hnRNP K (H-5): sc-365998 is recommended as a control antibody for monitoring of hnRNP K 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, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Lambda Phosphatase: sc-200312A 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 hnRNP K gene expression knockdown using RT-PCR Primer: hnRNP K (m)-PR: sc-38283-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.