SANTA CRUZ BIOTECHNOLOGY, INC.

symplekin siRNA (h): sc-97297

BACKGROUND

Symplekin, also known as SYMPK, SPK or SYM, is a regulatory protein that localizes to the nucleus, cytoplasm, cytoskeleton and cell junction. Expressed in a variety of tissues including testis, pancreas, stomach, liver and fetal brain, symplekin functions to mediate polyadenylation events and promote gene expression. Symplekin is thought to form a high molecular weight complex (called the tight junction complex) with proteins that are involved in polyadenylation and, once in this complex, may serve as a scaffold that recruits regulatory proteins to polyadenylation sites. In addition, symplekin functions independently and plays a role in the 3'-end maturation of histone mRNAs. Defects in the gene encoding symplekin are associated with hepa-tocellular carcinoma, suggesting a possible role for symplekin in tumor formation. Symplekin is expressed as two isoforms due to alternative splicing events.

REFERENCES

1. Keon, B.H., et al. 1996. Symplekin, a novel type of tight junction plaque protein. J. Cell Biol. 134: 1003-1018.
2. Ueki, K., et al. 1997. Chromosomal localization to 19q13.3, partial genomic structure and 5' cDNA sequence of the human symplekin gene. Somat. Cell Mol. Genet. 23: 229-231.
3. Takagaki, Y. and Manley, J.L. 2000. Complex protein interactions within the human polyadenylation machinery identify a novel component. Mol. Cell. Biol. 20: 1515-1525.
4. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 602388. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
5. Langbein, L., et al. 2003. Tight junction-related structures in the absence of a lumen: Occludin, claudins and tight junction plaque proteins in densely packed cell formations of stratified epithelia and squamous cell carcinomas. Eur. J. Cell Biol. 82: 385-400.
6. Barnard, D.C., et al. 2004. Symplekin and xGLD-2 are required for CPEB-mediated cytoplasmic polyadenylation. Cell 119: 641-651.

CHROMOSOMAL LOCATION

Genetic locus: SYMPK (human) mapping to 19q13.32.

PRODUCT

symplekin 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see symplekin shRNA Plasmid (h): sc-97297-SH and symplekin shRNA (h) Lentiviral Particles: sc-97297-V as alternate gene silencing products.

For independent verification of symplekin (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-97297A, sc-97297B and sc-97297C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

symplekin siRNA (h) is recommended for the inhibition of symplekin 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 µM in 66 µl. 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

symplekin (G-6): sc-398897 is recommended as a control antibody for monitoring of symplekin 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 symplekin gene expression knockdown using RT-PCR Primer: symplekin (h)-PR: sc-97297-PR (20 µl). 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.

PROTOCOLS

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