SANTA CRUZ BIOTECHNOLOGY, INC.

ESX1 siRNA (m): sc-77290

BACKGROUND

ESX1 was originally identified as a regulator of mouse embryogenesis. In mice, it is primarily expressed in placenta and testis where it functions in placenta/fetus development and spermatogenesis, respectively. In human cell lines, ESX1 has been elucidated as a paired-like homeoprotein that is proteolytically processed into N-terminal and C-terminal fragments. The N-terminal ESX1 fragment, which contains the homeodomain, localizes to the nucleus and represses mRNA transcription from the K-ras gene. A gain-of-function mutation of the K-ras gene is one of the most common genetic changes in human tumors. Therefore, ESX1 is implicated as a therapeutic target in the treatment of human cancers that have oncogenic K-Ras mutations.

REFERENCES

1. Li, Y., Lemaire, P. and Behringer, R.R. 1997. ESX1, a novel X chromosome-linked homeobox gene expressed in mouse extraembryonic tissues and male germ cells. Dev. Biol. 188: 85-95.
2. Li, Y. and Behringer, R.R. 1998. ESX1 is an X-chromosome-imprinted regulator of placental development and fetal growth. Nat. Genet. 20: 309-311.
3. Yan, Y.T., Stein, S.M., Ding, J., Shen, M.M. and Abate-Shen, C. 2000. A novel PF/PN motif inhibits nuclear localization and DNA binding activity of the ESX1 homeoprotein. Mol. Cell. Biol. 20: 661-671.
4. Yanagihara, M., Ishikawa, S., Naito, M., Nakajima, J., Aburatani, H. and Hatakeyama, M. 2005. Paired-like homeoprotein ESXR1 acts as a sequence-specific transcriptional repressor of the human K-Ras gene. Oncogene 24: 5878-5887.
5. Yeh, Y.C., Yang, V.C., Huang, S.C. and Lo, N.W. 2005. Stage-dependent expression of extra-embryonic tissue-spermatogenesis-homeobox gene 1 (ESX1) protein, a candidate marker for X chromosome-bearing sperm. Reprod. Fertil. Dev. 17: 447-455.
6. Wang, X. and Zhang, J. 2007. Rapid evolution of primate ESX1, an X-linked placenta- and testis-expressed homeobox gene. Hum. Mol. Genet. 16: 2053-2060.

CHROMOSOMAL LOCATION

Genetic locus: Esx1 (mouse) mapping to X F1.

PRODUCT

ESX1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ESX1 shRNA Plasmid (m): sc-77290-SH and ESX1 shRNA (m) Lentiviral Particles: sc-77290-V as alternate gene silencing products.

For independent verification of ESX1 (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-77290A, sc-77290B and sc-77290C.

RESEARCH USE

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

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

ESX1 siRNA (m) is recommended for the inhibition of ESX1 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 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

ESX1 (B-9): sc-365740 is recommended as a control antibody for monitoring of ESX1 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 ESX1 gene expression knockdown using RT-PCR Primer: ESX1 (m)-PR: sc-77290-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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