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SANTA CRUZ BIOTECHNOLOGY, INC.

Ythdc1 siRNA (m): sc-155421

BACKGROUND

YT521-B (YTH domain-containing protein 1), also known as YT521, is a 727 amino acid nuclear protein that localizes to the novel subnuclear structure of YT bodies and is the human homolog of the mouse gene, Ythdc1. Ubiquitously expressed, YT521-B may be part of a signal transduction pathway that influences splice site selection. YT521-B shuttles between the nucleus and cytosol, where it can be phosphorylated by c-Src or Fyn. Tyrosine phosphorylation by c-Abl causes dispersion of YT521-B from YT bodies to the nucleoplasm. Tyrosine phosphorylation also promotes sequestration of YT521-B in an insoluble nuclear form, which abolishes the ability of YT521-B to change alternative splice sites. YT521-B is considered to be a candidate for a role in a gene expression model of the pathogenesis of EDMD (Emery-Dreifuss muscular dystrophy), a type of muscular dystrophy primarily affecting voluntary muscles. YT521-B exists as two isoforms due to atlernative splicing events.

REFERENCES

1. Imai, Y., et al. 1998. Cloning of a gene, YT521, for a novel RNA splicing-related protein induced by hypoxia/reoxygenation. Brain Res. Mol. Brain Res. 53: 33-40.
2. Hartmann, A.M., et al. 1999. The interaction and colocalization of Sam68 with the splicing-associated factor YT521-B in nuclear dots is regulated by the Src family kinase p59(fyn). Mol. Biol. Cell 10: 3909-3926.
3. Nayler, O., et al. 2000. The ER repeat protein YT521-B localizes to a novel subnuclear compartment. J. Cell Biol. 150: 949-962.
4. Stoss, O., et al. 2001. The STAR/GSG family protein rSLM-2 regulates the selection of alternative splice sites. J. Biol. Chem. 276: 8665-8673.
5. Stoilov, P., et al. 2002. YTH: a new domain in nuclear proteins. Trends Biochem. Sci. 27: 495-497.
6. Wilkinson, F.L., et al. 2003. Emerin interacts *in vitro* with the splicing-associated factor, YT521-B. Eur. J. Biochem. 270: 2459-2466.
7. Rafalska, I., et al. 2004. The intranuclear localization and function of YT521-B is regulated by tyrosine phosphorylation. Hum. Mol. Genet. 13: 1535-1549.

CHROMOSOMAL LOCATION

Genetic locus: Ythdc1 (mouse) mapping to 5 E1.

PRODUCT

Ythdc1 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 Ythdc1 shRNA Plasmid (m): sc-155421-SH and Ythdc1 shRNA (m) Lentiviral Particles: sc-155421-V as alternate gene silencing products.

For independent verification of Ythdc1 (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-155421A, sc-155421B and sc-155421C.

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

Ythdc1 siRNA (m) is recommended for the inhibition of Ythdc1 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

YT521-B (8): sc-136428 is recommended as a control antibody for monitoring of Ythdc1 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 Ythdc1 gene expression knockdown using RT-PCR Primer: Ythdc1 (m)-PR: sc-155421-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.