

p35 siRNA (h): sc-36153

BACKGROUND

Cyclin dependent kinase-5 (Cdk5), a key regulator of cell cycle progression, was originally isolated on the basis of its structural homology to Cdc2, a well-characterized regulator of cell cycle progression. Although Cdk5 is expressed at the highest level in the brain of adult mice, intermediate levels in testis and low or undetectable levels in all other tissues, brain is the only tissue from which Cdk5 can be isolated as an active kinase. These findings may be explained by the cloning and characterization of a Cdk5 regulatory subunit, designated p35. p35 displays a neuronal cell-specific pattern of expression, physically associates with Cdk5 and activates Cdk5 enzymatic activity. p35 is also expressed in many tissues in a truncated form, designated p25.

REFERENCES

1. Murray, A.W. and Kirschner, M.W. 1989. Dominoes and clocks: the union of two views of the cell cycle. *Science* 246: 614-621.
2. Nurse, P. 1990. Universal control mechanism regulating onset of M-phase. *Nature* 344: 503-508.
3. Pines, J. and Hunter, T. 1990. p34cdc2: the S and M kinase? *New Biol.* 2: 389-401.
4. Draetta, G. 1990. Cell cycle control in eukaryotes: molecular mechanisms of cdc2 activation. *Trends Biochem. Sci.* 15: 378-383.
5. Meyerson, M., et al. 1992. A family of human cdc2-related protein kinases. *EMBO J.* 11: 2909-2917.
6. Tsai, L.H., et al. 1994. p35 is a neural-specific regulatory subunit of cyclin-dependent kinase 5. *Nature* 371: 419-423.

CHROMOSOMAL LOCATION

Genetic locus: CDK5R1 (human) mapping to 17q11.2.

PRODUCT

p35 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see p35 shRNA Plasmid (h): sc-36153-SH and p35 shRNA (h) Lentiviral Particles: sc-36153-V as alternate gene silencing products.

For independent verification of p35 (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-36153A, sc-36153B and sc-36153C.

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

p35 siRNA (h) is recommended for the inhibition of p35 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 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

p35 (G-7): sc-518015 is recommended as a control antibody for monitoring of p35 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 p35 gene expression knockdown using RT-PCR Primer: p35 (h)-PR: sc-36153-PR (20 μ l, 518 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Tripathi, B.K., et al. 2015. The Cdk5 activator P39 specifically links muskellin to myosin II and regulates stress fiber formation and actin organization in lens. *Exp. Cell Res.* 330: 186-198.

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.