

Dynactin 2 siRNA (h): sc-95708

BACKGROUND

Dynactin is a multisubunit complex that functions as a binding partner for the Dynein microtubule motor. Dynactin-Dynein binding may be required for most, if not all, cytoplasmic Dynein-driven activities and is thought to contribute to the functional diversity of Dynein. Dynactin 2, also known as DCTN2, Dynamin or DCTN50, is a peripheral membrane protein that is one of many subunits in the Dynactin complex. Like other Dynactin subunits, Dynactin 2 mediates Dynein-organelle binding and helps to regulate chromosome alignment during prometaphase and spindle organization during mitosis. Overexpression of Dynactin 2 disrupts the Dynactin-Dynein complex, thus inhibiting retrograde axonal transport and causing motor neuron degeneration. Additionally, overexpression of Dynactin 2 may disrupt the cell cycle and lead to osteosarcoma, suggesting a possible role for Dynactin 2 in carcinogenesis.

REFERENCES

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4. Karki, S., et al. 2000. A dynactin subunit with a highly conserved cysteine-rich motif interacts directly with Arp1. *J. Biol. Chem.* 275: 4834-4839.
5. Hoogenraad, C.C., et al. 2001. Mammalian Golgi-associated Bicaudal-D2 functions in the dynein-dynactin pathway by interacting with these complexes. *EMBO J.* 20: 4041-4054.
6. LaMonte, B.H., et al. 2002. Disruption of dynein/dynactin inhibits axonal transport in motor neurons causing late-onset progressive degeneration. *Neuron* 34: 715-727.
7. Uetake, Y., et al. 2004. Interaction of Cep135 with a p50 dynactin subunit in mammalian centrosomes. *Cell Motil. Cytoskeleton* 58: 53-66.
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CHROMOSOMAL LOCATION

Genetic locus: DCTN2 (human) mapping to 12q13.3.

PRODUCT

Dynactin 2 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 Dynactin 2 shRNA Plasmid (h): sc-95708-SH and Dynactin 2 shRNA (h) Lentiviral Particles: sc-95708-V as alternate gene silencing products.

For independent verification of Dynactin 2 (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-95708A, sc-95708B and sc-95708C.

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

Dynactin 2 siRNA (h) is recommended for the inhibition of Dynactin 2 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

Dynactin 2 (G-4): sc-393389 is recommended as a control antibody for monitoring of Dynactin 2 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 Dynactin 2 gene expression knockdown using RT-PCR Primer: Dynactin 2 (h)-PR: sc-95708-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.

PROTOCOLS

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