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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



Dynamin I/II shRNA (h) Lentiviral Particles: sc-43736-V

BACKGROUND

Members of the dynamin family, including Dynamin I and Dynamin II, are GTPase, microtubule-associated proteins which are involved in endocytosis, synaptic transmission and neurogenesis. Dynamin I is localized to the central nervous system, while Dynamin II exhibits ubiquitous distribution with highest expression found in testis. Both dynamin proteins contain SH3 and proline-rich domains that mediate interactions between the dynamins and effectors of their GTPase activity. The interactions with these effectors, which include microtubules, acidic phospholipids and SH3 domain-containing proteins, are required for rapid endocytosis. Dynamin I appears to be recruited to clathrin coated pits by SH3 domain interaction with amphiphysin, a protein highly expressed in brain.

REFERENCES

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5. Okamoto, P.M., et al. 1997. Role of the basic, proline-rich region of dynamin in Src homology 3 domain binding and endocytosis. *J. Biol. Chem.* 272: 11629-11635.
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7. Grabs, D., Slepnev, V.I., Songyang, Z., David, C., Lynch, M., Cantley, L.C. and de Camilli, P. 1997. The SH3 domain of amphiphysin binds the proline-rich domain of dynamin at a single site that defines a new SH3 binding consensus sequence. *J. Biol. Chem.* 272: 13419-13425.

CHROMOSOMAL LOCATION

Genetic locus: DNM1 (human) mapping to 9q34.11.

PRODUCT

Dynamin I/II shRNA (h) Lentiviral Particles are concentrated, transduction-ready viral particles containing a target-specific construct that encodes a 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Dynamin I/II siRNA (h): sc-43736 and Dynamin I/II shRNA Plasmid (h): sc-43736-SH as alternate gene silencing products.

APPLICATIONS

Dynamin I/II shRNA (h) Lentiviral Particles is recommended for the inhibition of Dynamin II expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Dynamin I/II (E-4): sc-390160 is recommended as a control antibody for monitoring of Dynamin I/II 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Dynamin II gene expression knockdown using RT-PCR Primer: Dynamin I/II (h)-PR: sc-43736-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.