

p-Dynamin I (E-9): sc-377563

BACKGROUND

Dynamin I is a GTPase enzyme required for the retrieval of synaptic vesicles after exocytosis and functions in endocytosis by stimulating assembly of invaginating synaptic vesicles. Dynamin I is phosphorylated in nerve terminals exclusively in the cytosolic compartment and *in vitro* by protein kinase C (PKC). The phosphorylation site in PKC-phosphorylated Dynamin I is a single site at Serine 795, which is located near a binding site for the SH3 domain of p85, the regulatory subunit of phosphatidylinositol 3-kinase. Dephosphorylation is required for synaptic vesicle retrieval, suggesting that phosphorylation affects the subcellular localization of Dynamin I.

REFERENCES

1. Koenig, J.H. and Ikeda, K. 1989. Disappearance and reformation of synaptic vesicle membrane upon transmitter release observed under reversible blockage of membrane retrieval. *J. Neurosci.* 9: 3844-3860.
2. Robinson, P.J. 1991. Dephosphin, a 96,000 Da substrate of protein kinase C in synaptosomal cytosol, is phosphorylated in intact synaptosomes. *FEBS Lett.* 282: 388-392.
3. Robinson, P.J., Sontag, J.M., Liu, J.P., Fykse, E.M., Slaughter, C., McMahon, H. and Sudhof, T.C. 1993. Dynamin GTPase regulated by protein kinase C phosphorylation in nerve terminals. *Nature* 365: 163-166.
4. Liu, J.P., Powell, K.A., Sudhof, T.C. and Robinson, P.J. 1994. Dynamin I is a Ca²⁺-sensitive phospholipid-binding protein with very high affinity for protein kinase C. *J. Biol. Chem.* 269: 21043-21050.
5. Powell, K.A., Valova, V.A., Malladi, C.S., Jensen, O.N., Larsen, M.R. and Robinson, P.J. 2000. Phosphorylation of dynamin I on Ser-795 by protein kinase C blocks its association with phospholipids. *J. Biol. Chem.* 275: 11610-11617.

CHROMOSOMAL LOCATION

Genetic locus: DNM1 (human) mapping to 9q34.11; Dnm1 (mouse) mapping to 2 B.

SOURCE

p-Dynamin I (E-9) is a mouse monoclonal antibody specific for an epitope mapping between amino acids 789-804 Ser 795 of Dynamin I of human origin.

PRODUCT

Each vial contains 200 µg IgG₁ kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-377563 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

STORAGE

Store at 4° C, ****DO NOT FREEZE****. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

APPLICATIONS

p-Dynamin I (E-9) is recommended for detection of Ser 795 phosphorylated Dynamin I of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

p-Dynamin I (E-9) is also recommended for detection of correspondingly phosphorylated Dynamin I in additional species, including canine, bovine and porcine.

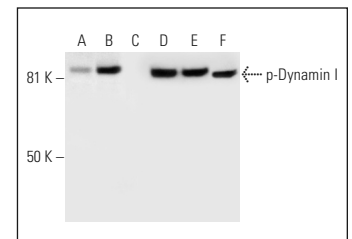
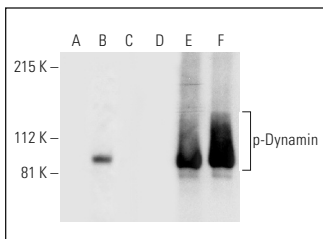
Suitable for use as control antibody for Dynamin I siRNA (h): sc-43737, Dynamin I siRNA (m): sc-35234, Dynamin I shRNA Plasmid (h): sc-43737-SH, Dynamin I shRNA Plasmid (m): sc-35234-SH, Dynamin I shRNA (h) Lentiviral Particles: sc-43737-V and Dynamin I shRNA (m) Lentiviral Particles: sc-35234-V.

Molecular Weight of p-Dynamin I: 100 kDa.

RECOMMENDED SUPPORT REAGENTS

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, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Lambda Phosphatase: sc-200312A and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) 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.

DATA



Western blot analysis of Dynamin I phosphorylation in non-transfected (A,D), untreated human Dynamin I transfected: sc-117282 (B,E) and lambda protein phosphatase (sc-200312A) treated human Dynamin I transfected: sc-117282 (C,F) 293T whole cell lysates. Antibodies tested include p-Dynamin I (E-9): sc-377563 (A,B,C) and Dynamin I (D5): sc-12724 (D,E,F).

Western blot analysis of Dynamin I phosphorylation in untreated (A,D), Ser/Thr Phosphorylation Induction Cocktail (sc-362324) treated (B,E) and Ser/Thr Phosphorylation Induction Cocktail (sc-362324) and lambda protein phosphatase (sc-200312A) treated (C,F) SH-SY5Y whole cell lysates. Antibodies tested include p-Dynamin I (E-9): sc-377563 (A,B,C) and Dynamin I (D5): sc-12724 (D,E,F).

RESEARCH USE

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