

p-Vimentin (Ser 83): sc-16674

BACKGROUND

Phosphorylation of Vimentin induces disassembly of Vimentin intermediate filaments *in vivo* and *in vitro*. Binding of 14-3-3 depends on Vimentin phosphorylation and requires the phosphopeptide binding domain of 14-3-3, which is an amino terminal head domain consisting of amino acids 1-96. Phosphorylated Vimentin sequesters 14-3-3 and limits its availability to other target proteins, which can affect intracellular signaling processes that require 14-3-3. The amino-terminal domain of Vimentin is the target site for several protein kinases, including Rho kinase and PKC. Ser 38 and Ser 71 of Vimentin are the major sites of phosphorylation by Rho kinase. The disruption of sub-cellular compartmentalization of interphase cells leads to PKC-mediated phosphorylation of Vimentin. Thus, targeting of activated PKC, coupled with the reorganization of intracellular membranes, which contain phospholipids essential for activation, leads to the mitosis-specific phosphorylation of Vimentin.

REFERENCES

1. Takai, Y., Ogawara, M., Tomono, Y., Moritoh, C., Imajoh-Ohmi, S., Tsutsumi, O., Taketani, Y. and Inagaki, M. 1996. Mitosis-specific phosphorylation of Vimentin by protein kinase C coupled with reorganization of intracellular membranes. *J. Cell Biol.* 133: 141-149.
2. Goto, H., Kosako, H., Tanabe, K., Yanagida, M., Sakurai, M., Amano, M., Kaibuchi, K. and Inagaki, M. 1998. Phosphorylation of Vimentin by Rho-associated kinase at a unique amino-terminal site that is specifically phosphorylated during cytokinesis. *J. Biol. Chem.* 273: 11728-11736.
3. Tziviion, G., Luo, Z.J. and Avruch, J. 2000. Calyculin A-induced Vimentin phosphorylation sequesters 14-3-3 and displaces other 14-3-3 partners *in vivo*. *J. Biol. Chem.* 275: 29772-29778.
4. Nakamura, Y., Hashimoto, R., Amano, M., Nagata, K., Matsumoto, N., Goto, H., Fukusho, E., Mori, H., Kashiwagi, Y., Kudo, T., Inagaki, M. and Takeda, M. 2000. Localized phosphorylation of Vimentin by Rho-kinase in neuroblastoma N2a cells. *Genes Cells* 5: 823-837.
5. Gohara, R., Tang, D., Inada, H., Inagaki, M., Takasaki, Y. and Ando, S. 2001. Phosphorylation of Vimentin head domain inhibits interaction with the carboxyl-terminal end of α -helical rod domain studied by surface plasmon resonance measurements. *FEBS Lett.* 489: 182-186.

CHROMOSOMAL LOCATION

Genetic locus: VIM (human) mapping to 10p13; Vim (mouse) mapping to 2 A2.

SOURCE

p-Vimentin (Ser 83)-R is a rabbit polyclonal affinity purified antibody raised against a short amino acid sequence containing Ser 83 phosphorylated Vimentin of human origin.

STORAGE

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

PRODUCT

Each vial contains 200 μ g IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

Blocking peptide available for competition studies, sc-16674 P, (100 μ g peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% BSA).

APPLICATIONS

p-Vimentin (Ser 83) is recommended for detection of Ser 83 phosphorylated Vimentin of mouse, rat, human and *Xenopus laevis* origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), 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-Vimentin (Ser 83) is also recommended for detection of correspondingly phosphorylated Vimentin in additional species, including equine, canine, bovine and porcine.

Suitable for use as control antibody for Vimentin siRNA (h): sc-29522, Vimentin siRNA (m): sc-29523, Vimentin shRNA Plasmid (h): sc-29522-SH, Vimentin shRNA Plasmid (m): sc-29523-SH, Vimentin shRNA (h) Lentiviral Particles: sc-29522-V and Vimentin shRNA (m) Lentiviral Particles: sc-29523-V.

Molecular Weight of p-Vimentin: 57 kDa.

Positive Controls: HeLa + Calyculin A cell lysate: sc-2271.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (range: 1:2000-1:5000); Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), and Lambda Phosphatase: sc-200312A. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

SELECT PRODUCT CITATIONS

1. Daily, A., Monks, N.R., Leggas, M. and Moscow, J.A. 2010. Abrogation of microcystin cytotoxicity by MAP kinase inhibitors and N-acetyl cysteine is confounded by OATPIB1 uptake activity inhibition. *Toxicol* 55: 827-837.

RESEARCH USE

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

PROTOCOLS

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