3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

ProductInformation

Anti-ROCK-2
Developed in Rabbit, Affinity Isolated Antibody

Product Number **R 8653**

Product Description

Anti-ROCK-2 is developed in rabbit using a synthetic peptide corresponding to amino acids 1371-1388 located at the C-terminus of human ROCK-2, conjugated to KLH, as immunogen. This sequence is identical in rat, mouse and bovine ROCK-2 and is highly conserved in *Xenopus* ROCK-2. The antibody is affinity-purified using the immunizing peptide immobilized on agarose.

Anti-ROCK-2 recognizes ROCK-2 (160 kDa) by immunoblotting.

Actin cytoskeletal reorganization is essential for various cell activities, including motility, morphological change, adhesion and cytokinesis. Various extracellular stimuli induce changes in actin organization, through signaling pathways that link the external stimuli to the machinery controlling actin polymerization and organization. The Rho family of small GTPases, Rho, Rac and Cdc42 play a central role in regulating actin organization through downstream effectors, the activity of which is controlled by interactions with the active, GTP-bound forms of the Rho family.[1-2] Several Rho effector proteins have been described including citron, rhotekin, rhophilin, protein kinase N and Rho-associated kinases.[3-5] Rho-associated kinases (termed ROCK, ROK, or Rho-kinase) are serine/threonine kinases implicated in Rho-mediated actin reorganization such as formation of stress fibers and focal adhesions and smooth muscle contraction.[4-8] Two ROCK isoforms have been described, ROCK-1 (also referred to as $p160^{ROCK}$, ROCK-β, 160 kDa) and ROCK-2 (also referred to as ROCK-α, 150-160kDa) that share a structural similarity to myotonic dystrophy kinase.[4,5] Rho-kinases lead to an increase of myosin light chain (MLC) phosphorylation and thereby induce actomyosin-based contractility.[9,10] ROCK accumulates at the cleavage furrow during cytokinesis. In addition, a dominant negative form of ROCK increases multinuclei in mammalian cells. ROCK directly phosphorylates and activates LIM-kinase (LIMK), resulting in downstream phosphorylation and inaction of the actin-depolymerizing factor cofilin.[11-13]

Reagent

The product is provided as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as preservative.

Antibody Concentration: approx.2.0 mg/mL

Precautions and Disclaimer

Due to the sodium azide content a material safety data sheet (MSDS) for this product has been sent to the attention of the safety officer of your institution. Consult the MSDS for information regarding hazardous and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage freeze in working aliquots. Repeated freezing and thawing is not recommended. Storage in "frost-free" freezers is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

Product Profile

A working dilution of 2-4 µg/ml is determined by immunoblotting, using a whole cell extract of human acute T cell leukemia Jurkat cell line and of mouse fibroblasts NIH3T3 cell line. Staining of the ROCK-2 band in immunoblotting is specifically inhibited with the ROCK-2 immunizing peptide (human, amino acids 1371-1388).

A working dilution of 3-6 µg/ml is determined by immunofluorescence staining of mouse fibroblasts NIH3T3 cell line.

25-50 µg of the antibody can immunoprecipitate ROCK-2 protein from a Jurkat cell lysate.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working concentrations by titration test.

References

1. Hall, A., Science, **279**, 509-514 (1998).
2. Etienne-Manneville, S., and Hall, A., Nature, **420**, 629-635 (2002).
3. Watanabe, G, et al., Science, **271**, 645-648 (1996).
4. Ishizaki, T., et al., EMBO J., **15**, 1885-1893 (1996).
5. Leung, T., et al., J. Biol. Chem., **270**, 29051-29054 (1995).
6. Amano, M., et al., Exp. Cell Res., **261**, 44-51 (2000).
7. Amano, M., et al., Science, **275**, 1308-1311 (1997).
8. Leung, T., et al., Mol. Cell. Biol., **16**, 5313-5327 (1996).
9. Amano, M., et al., J. Biol. Chem., **271**, 20246-20249 (1996).
10. Kureishi, Y., et al., J. Biol. Chem., **272**, 12257-12260 (1997).
11. Maekawa, M., et al., Science, **271**, 895-898 (1999).
12. Ohashi, K., et al., J. Biol. Chem., **275**, 3577-3582 (2000).
13. Sumi, T., et al., J. Biol. Chem., **276**, 670-676 (2001).

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