

Product Information

Anti-Actopaxin

Developed in Rabbit
IgG Fraction of Antiserum

Product Number **A 1226**

Product Description

Anti-Actopaxin is developed in rabbit using a synthetic peptide corresponding to the N-terminal region of mouse actopaxin (amino acids 35-53), conjugated to KLH as immunogen. The sequence is identical in human actopaxin and has considerable homology (~70%) with β -parvin/affixin, but no homology with γ -parvin. Whole antiserum is fractionated and then further purified by ion-exchange chromatography to provide the IgG fraction of antiserum that is essentially free of other rabbit serum proteins.

Anti-Actopaxin, recognizes actopaxin (42 kDa). Applications include the detection of actopaxin by immunoblotting. Staining of actopaxin in immunoblotting is specifically inhibited with actopaxin immunizing peptide (mouse, amino acids 35-53).

Cell adhesion to the extracellular matrix (ECM) is an important process that controls cell morphology, proliferation, migration, differentiation and survival. Cell adhesion to the ECM and engagement of integrin molecules on the cell surface is accompanied by recruitment of multiple cytoskeletal and signaling proteins to focal adhesion sites.¹⁻⁴ These proteins link the cytoskeleton to the ECM and mediate signal transduction between the ECM and the intracellular compartment. Actopaxin (also termed α -parvin, CH-ILKBP) is a novel, ubiquitously expressed 42 kDa focal adhesion protein, involved in the regulation of cell adhesion.⁵⁻⁷ Actopaxin belongs to a family of actin-binding proteins that includes β -parvin/affixin and γ -parvin.^{7, 8} It contains two calponin-homology (CH) domains and a paxillin-binding subdomain (PBS).

Actopaxin binds directly to both F-actin and to paxillin LD motifs LD1 and LD4. In addition, actopaxin and the actopaxin/ parvin family member β -parvin/affixin, associates directly with the serine/threonine integrin-linked kinase (ILK). Actopaxin binds via the CH2 domain to the C-terminus of ILK, indicating that paxillin and ILK binding sites are distinct.⁹ Actopaxin localizes to focal adhesions but is not found along the length of the associated actin-rich stress fibers. Similar to paxillin it is absent from actin-rich cell-cell adherens junctions. Actopaxin colocalizes with paxillin to rudimentary focal complexes at the leading edge of migrating cells. Paxillin is necessary for actopaxin recruitment to focal adhesions. An actopaxin PBS mutant incapable of binding paxillin *in vitro* cannot target to focal adhesions when expressed in fibroblasts. In addition, ectopic expression of a PBS mutant and/or the C-terminus of actopaxin in HeLa cells results in substantial reduction in cell adhesion/ spreading on collagen. Actopaxin is phosphorylated during mitosis and is a substrate for cyclin B/cdc2 kinase.¹⁰ As cells progress from mitosis to G₁ there is an adhesion-dependent dephosphorylation of actopaxin, preceding cell spreading and reformation of focal adhesions. These studies suggest an important role for actopaxin in integrin-dependent remodeling of the actin cytoskeleton during cell adhesion, motility and cell division.

Reagent

Anti-Actopaxin is supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide.

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 hazards and safe handling practices.

Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For prolonged storage, freeze in working aliquots at -20 °C. Repeated freezing and thawing is not recommended. Storage in frost-free freezers is also not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilutions should be discarded if not used within 12 hours.

Product Profile

For immunoblotting, a minimum working antibody dilution of 1:3,000 is recommended using a whole cell extract of the human endothelial ECV304 cell line.

Note: In order to obtain the best results using various techniques and preparations, we recommend determining the optimal working dilutions by titration.

References

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ER/KAA 10/02

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