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### Antibodies to Rho GTPase-activating protein RICH2

Rho GTPase-activating protein RICH2 antibodies. Catalog # RICH2-201AP, FITC-RICH2, P-RICH2 and PC-RICH2.

Accession # NP\_778168 and Alternate Nomenclature: Rho-type GTPase-activating protein RICH2; RP23-219K7.2; AU040829; AI840762

RICH2, Rho GTPase-activating protein interacting with CIP homologue protein 2, is a member Rho GTPase activating protein RICH family. This family of protein is one of the major regulators of Rho GTPases in all eukaryotes (1). In addition to their role as specific negative regulators of Rho protein signaling pathways, they are crucial in the differentiation, growth and organization of cell cytoskeleton, neuronal development and synaptic functions (2). RICH2 is located at mouse chromosome 11B3 and as a GTPase activator for the Rho-type GTPases, it converts them to an inactive GDP-bound state. Recent reports have identified RICH2 as a GTPase activator for Cdc42 and Rac1 (3). RICH2 and different isoforms of RICH1 share two conserved domains, an N-terminal BAR domain and Rho/Rac/cdc42 GAP domain. The BAR domain of RICH2 binds membranes and is capable of inducing membrane tabulation (4). As a member of RICH family of proteins, RICH2 plays a crucial role in linking CD317/tetherin to the apical actin network (5).

Knocking down the expression of RICH2 in cells usually results in loss of apical actin network. These cells exhibited similar characteristics that were seen in the CD317 knockdown cells (5). These characteristics include subsequent loss of apical microvilli, increase in actin bundles at the basal surface, reduction in cell height without any loss of tight junctions or transepithelial resistance or any change in the distribution of apical and basolateral marker proteins. Overexpression of RICH2 leads to a loss of Ras-induced membrane ruffles (3). RICH2 is highly expressed in brain and liver. It is approximately a 125kDa protein (5).

The RICH2-selective antibodies were generated against purified RICH2 protein. RICH2-antibodies are affinity purified over immobilized antigen based chromatography, and the purified immunoglobulins are stabilized in antibody stabilization buffer. FabGennix Int. Inc., will also provide limited quantities of antigenic blocking protein for RICH2 -201AP antibody. Antibodies to RICH2 (RICH2 -201AP) will label ~125kDa protein in Western blot positive control samples for RICH2 and several other tissues. FabGennix Inc. will also conjugate antibodies with secondary enzymes (alk-Pase or HRP) or fluorescent probes upon request at nominal cost.

Catalog #	Host Species	Nature	Cross reactivity	Quantity	Volume
RICH2-201 AP	Rabbit	Affinity purified RICH2 antibodies	M, R, H, others	100 ug	200ul
FITC- RICH2	Rabbit	FITC-conjugated RICH2 antibody	M, R, H, others	100ug	200ul
P- RICH2	n/a	Antigenic blocking peptide for RICH2 -201AP	n/a	250 ug	100ul
PC- RICH2	n/a	Western blotting positive control for RICH2	n/a	For 5 appl	Inquire

R = rat; M = mouse; H = human; C = chicken; monk = monkey; \* not all variants are labeled equally

**Immunogen:** Synthetic peptides corresponding to unique epitope on RICH2. The peptide was taken from N-terminal 1-60 amino acids. The RICH2 peptide was covalently modified post-synthetically covalently modified to achieve desired antigenicity.

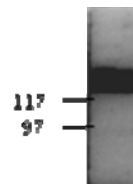
**Concentration:** RICH2 -201AP: IgG concentration 0.64-0.72 mg/ml in antibody stabilization buffer.

**Applications:** Antibody RICH2 -201AP is ideal for WB and ELISA applications, other applications have not been tested. The species cross reactivity for these antibodies have not been examined fully. The dilutions for this antibody is for reference only, investigators are expected to determine the optimal conditions for specific assay. WB: > 1:500; IMM & i.p pull-down assays: n.d; IHC n.d. ELISA <1:10,000. Application of this antibody in protocols not listed here does not necessarily exclude its use in such procedures.

**Reactivity:** This antibody detects a single band of approximately 125kDa in PC- RICH2 samples.

**Protocols:** Standard protocol for various applications (WB; IMM and IHC) of this antibody is provided with the product specification sheet, however, FabGennix Inc., strongly recommends investigators to optimize conditions for use of this antibody in their laboratories.

**Form/Storage:** The antiserum is supplied in antibody stabilization buffer. The affinity-purified antibodies are isolated on immobilized antigen-affinity column and supplied as stabilized product. Store at -20°C for long-term storage. FabGennix Inc. does not recommend storage of very dilute antibody solutions unless they are prepared in specially formulated multi use antibody dilution buffer (Cat # DiluOBuffer). Working solutions of antibodies in DiluOBuffer should be filtered through 0.45um filter after every use for long-term storage.



Western Blot of RICH2. The RICH2 antibody (Cat # RICH2-101AP) at 1:500 in diluOBuffer was probed with PC-RICH2 sample. MW of RICH2 is approximately 125kDa

**Notes:** Briefly centrifuge to collect liquid, heat or boil PC-RICH2 tube for 1-2 minutes to dissolve any precipitate before use. This product is "ready-to-use" for electrophoresis. After thawing store at room temperature, Repeated freezing and thawing may result in appearance of higher MW immunoreactive bands.

**New Reagents:** Now you can recycle your western blots (nitrocellulose, supported membranes and PVDF membranes) by using our StripOBuffer (Cat FGI-1989). Each stripping is guaranteed to give better signal (up to 8 stripping). No strong pungent smell of reducing agents or heating required. Block in 5X diluOBuffer and you are ready for blotting with a new antibody

**References:**

1. Furuta B, et al. Identification and functional characterization of nadrin variants, a novel family of GTPase activating protein for rho GTPases. J. Neurochem. 2002; 82:1018-1028.
2. Moon SY, Zheng Y. Rho GTPase-activating proteins in cell regulation. Trends in Cell Biology. January 2003; 13(1):13-22.
3. Richnau N, Aspenstrom P. Rich, a Rho GTPase-activating protein domain containing protein involved in signaling by Cdc42 and Rac1. J. Biol. Chem. 2001; 276:35060-35070.
4. Richnau N, et al. RICH-1 has a BIN/Amphiphysin/Rvsp domain responsible for binding to membrane lipids and tabulation of liposomes. Biochem. Biophys. Res. Commun. 2004; 320:1034-1042.
5. Rollason R, et al. A CD317/tetherin-RICH2 complex plays a critical role in the organization of the subapical actin cytoskeleton in polarized epithelial cells. J Cell Biol. 9 March 2009; 184(5):721-736.

\* For users who may require large amounts of RICH2-201AP, please enquire about bulk material discounts.  
This Product is for Research Use Only and is NOT intended for use in humans or clinical diagnosis.

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