



**FabGennix Inc.**  
INTERNATIONAL

New Item  
New Item

Customer Service: 1-800 786 1236  
Technical Service: 214 387 8105  
Fax: 214 387 0870  
[Info@fabgennix.com](mailto:Info@fabgennix.com)  
[www.fabgennix.com](http://www.fabgennix.com)

### Non-Receptor Tyrosine kinases selective antibodies

#### Anti-phospho-ACK1 antibodies (PACK-140AP)

ACK1 (activated Cdc42-associated kinase 1) is a nonreceptor tyrosine kinase and the only tyrosine kinase known to interact with Cdc42. ACK1 phosphorylates and activates the guanine nucleotide exchange factor Dbl, which in turn directs the Rho family GTP-binding proteins (1). ACK1 interacts most strongly with the SH3 domains of Src family kinases (Src or Hck) via its C-terminal proline-rich domain. The ACK1 interacts most strongly with the SH3 domains of Src family kinases (Src or Hck) by its C-terminal proline rich domain. ACK1 also acts as a regulator of Dbl, which in turn activates Rho family proteins. There are at least 2 ACK1 associated proteins (Cdc42 and Grb) that are required for ACK1-dependent EGF signaling in the cell. The central motif in ACK1 has a clathrin adaptor that co-localizes ACK1 with in the clathrin containing vesicles that are involved in the trafficking of receptor mediated endocytosis (2).

The ACK family of non-receptor tyrosine kinase (ACK1 and ACK2) specifically associate with Cdc42 to modulate its activity in several signaling pathways. ACK1 acts as a mediator of EGF signal to Rho-family GTP-binding proteins (3). The activation of ACK1 involves auto-phosphorylation at Tyrosine 284, upon phosphorylation, the ACK1 activity is significantly enhanced. Tyrosine substitution at 284 by Phenylalanine (Y284F) and expression in COS-7 cells exhibit dramatically reduced levels of tyrosine phosphorylation (1). ACK2 mediates cell adhesion signals initiated by integrins  $\beta 1$  in a Cdc-dependent manner (4). There are three structural domains associated with ACK1 protein, a N-terminal tyrosine kinase domain, SH3 domain, followed by a Cdc42/Rac interactive binding (CRIB) domain and a proline rich region near C-terminal end. ACK2 has been shown to be activated by Cdc42 binding to the CRIB region (4).

The ACK1 protein is a approximately 118-120kDa (1036 amino acids) protein. The Anti-ACK1-selective antibody was generated against a peptide from the tyrosine kinase domain that is unique to ACK1 protein. The affinity purified mono-specific polyclonal antibody strongly labels a 118-120kDa protein in ACK1 western blot positive controls. *FabGennix Inc.* will also conjugate antibodies with fluorescent probes upon request at extra charge. *FabGennix Inc.* also provides antibodies against proteins that are involved in ACK1 signaling including ACK2, phosphor-ACK1, Herceptins (Erbs) and others. *FabGennix Inc.* employs cyclic peptide methodology for generating antibodies, which results in higher titer and specificity (6). *FabGennix, Inc.*, will also provide Western blot positive controls for phosphor and non-phospho ACK1 in ready-to-use buffer for easy identification of respective proteins. Limited quantities of antigens are also available. Please enquire for their availability before ordering.

Catalog #	Host Species	Nature	Cross reactivity	Quantity	Volume
PACK-140AP	Rabbit	Affinity purified Phospho ACK1 Antibodies	R, M, H, monk	100/ g	175-200ul
P-ACK1	n/a	Non-Phospho blocking peptide	R, M, H, monk	250 ug	100 ul
P-PACK1	n/a	Antigenic blocking phospho peptide	R, M, H, monk	250 ug	100 ul
PAC-PACK1	n/a	Western blot positive control for Phospho-ACK1	R, M, H, monk	5 appl	200 ul
PC-ACK1	n/a	Western blot positive control for ACK1	R, M, H, monk	5 appl	200 ul

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

**Immunogen:** Synthetic phospho-tyrosine containing peptide unique to ACK1 protein that is known to phosphorylate by tyrosine kinase.

**Concentration:** PACK-140AP IgG concentration 0.65-1.25 mg/ml.

**Applications:** Antibody ACK-101AP is ideal for IMM and WB. ACK-101AP has not been tested in other applications. The dilutions for these antibodies are for reference only, investigators are expected to determine the optimal conditions for specific assay. WB; 1:500; IMM & i.p pull-down assays:> 1:200 (1 ul/250 ug protein extracts)

**Reactivity:** This antibody detects a single 120kDa ACK1 protein in PC-PACK1 samples. The antibody also reacts to a 110-120 kDa protein in various breast cancer cell lines.

**Protocols:** Standard protocol for various applications (WB, IMM and IHC) of this antibody is provided with the product specification sheet, however, *FabGennix Int. Inc.*

**Form/Storage:** The antiserum is supplied in antibody stabilization buffer with 0.02% sodium azide. For long-term storage of antibodies, store at -20°C. *FabGennix Int. Inc.* does not recommend storage of very dilute antibody solutions unless they are prepared in specially formulated multi use antibody dilution buffer (Cat # DilUbuffer). Working solutions of antibodies in DilUbuffer should be filtered through 0.45u filter after every use for long-term storage.

#### References:

1. Kato-Stankiewicz J, Ueda S, Kataoka T, Kaziro Y, Satoh T. *Biochem Biophys Res Commun.* 2001 8;284:470-7)
2. Teo M, Tan L, Lim L, Manser E. *J Biol Chem.* 2001 May 25;276(21):18392-8. Epub 2001 Feb 27
3. Kato-Stankiewicz et al., *Biochem. Biophys. Res. Comm.* 284, 470-477, 2004.
4. Yang W., et. Al., *J. Biol. Chem.* 274, 8524-8530, 1999.

Note: 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 is required.

\*For users who may require large amounts of PACK-140AP, please enquire about bulk material discounts.  
This Product is for Research Use Only and is NOT intended for use in humans or clinical diagnosis.

082705-0020SF1001Z-rev10.00

**FabGennix Inc.**  
INTERNATIONAL

5850 Town and Country Blvd. Suite 301. Frisco, TX 75034

Customer service: 1800 786 1236; Technical Support: 214 387 8105