

FAK [pY576] Rabbit Polyclonal Antibody

Store at 2° C to 8° C (short-term), or -20° C (long-term)

Catalog Number: 44-652G

Pub. No. MAN0010631 Rev. 1.00

Clonality: Polyclonal	Quantity: 10 mini-blots	Volume : 100 μL
Host/Class: Rabbit IgG	Reactivity: Human FAK [pY576]	Predicted Reactivity: Mouse, Chicken, Rat, Frog

Product description

Focal Adhesion Kinase (FAK) is a widely expressed cytoplasmic non-receptor protein tyrosine kinase. It is a substrate for Src and a key element in growth factor and integrin signaling. FAK plays a central role in cell spreading, differentiation, migration, cell death and acceleration of the G1 to S phase transition of the cell cycle. FAK is regulated through phosphorylation at multiple tyrosine and serine residues. Tyrosines 576 and 577 in the activation loop of the catalytic domain results in maximal activity of FAK, but the two sites appear to be regulated by different stimuli.

Product specifications

Immunogen:	A chemically synthesized phosphopeptide derived from the region of human FAK containing tyrosine 861.
Purification:	Antibody negatively preadsorbed using a non- phosphopeptide then purified by epitope-specific affinity chromatography
Apparent MW:	125 kDa
Gene ID:	5747
Protein Accession No.:	Q05397
Sequence Homology:	Mouse, Rat, Chicken, Frog
Lot:	See product label

Product applications

The antibody has been used in western blotting (1:1000 dilution) and immunostaining applications. Other applications may work but have not been tested.

Because conditions may vary, it is recommended that each investigator determine the optimal amount of antibody to be used for each application.

Storage and handling

Store the antibody at 2°C to 8°C for up to 1 week, or apportion into working aliquots and keep at -20°C for long-term storage. Avoid repeated freezing and thawing.

Stability

When stored as instructed, expires one year from date of receipt unless otherwise indicated on the Certificate of Analysis.

1 kDa 2 3 4 5 6 250 150 100 75 50 None (-) None (+) Generic pY FAK [pY5%] NH Pyk2 [pY579] FAK [pY576]

Figure 1 Antibody-Peptide Competition.

Extracts of primary CEF cells either mock transfected (lane 1), or transfected with a FAK vector (lanes 2–6), plated on fibronectin, and treated with 50 µM vanadate for 16 hours were resolved on a 10% Tris-glycine gel and transferred to PVDF. The membrane was blocked with 5% BSA-TBST for one hour at room temperature then incubated with the FAK [pY576] antibody for 2 hrs at room temperature in 1% BSA-TBST, following prior incubation with: no peptide (lanes 1, 2), a non-phosphorylated peptide corresponding to the immunogen (lane 3), a generic phosphotyrosine-containing peptide (lane 4), a phosphopeptide derived from the corresponding region of Pyk2 (lane 5), or the phosphopeptide immunogen (lane 6). The blots were developed using chemiluminescence (ECL) method with a goat F(ab')² antirabbit IgG HRP conjugate (Cat. no. ALI4404).

Only the phosphopeptide corresponding to FAK [pY861] blocks the antibody signal (lane 6) demonstrating the specificity of the antibody.

Positive controls used

Primary chicken embryo fibroblasts (CEF) expressing FAK protein and plated on fibronectin, or A459 whole cell lysate.

For Research Use Only. Not for use in diagnostic procedures

Storage buffer

Dulbecco's phosphate buffered saline (without Mg²⁺ and Ca²⁺), pH 7.3 (+/– 0.1), 50% glycerol with 1.0 mg/mL BSA (IgG, protease free) as a carrier, and 0.05% sodium azide.

CAUTION! Sodium azide is extremely toxic and may react with lead and copper plumbing to form highly explosive metal azides. Properly dispose of solutions containing sodium azide. Read the Safety Data Sheet (SDS) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves. SDSs are available at www.lifetechnologies.com/support.

References

- 1. Dunty, J., et al. (2004) FERM domain interaction promotes FAK signaling. *Mol. Cell. Biol.* 24(12):5353-5368.
- Shikata, Y., et al. (2003) Involvement of site-specific FAK phosphorylation in sphingosine-1 phosphate- and thrombininduced focal adhesion remodeling: role of Src and GIT. *FASEB J*. 17(15):2240-2249.
- 3. Cary, L.A., et al. (2002) SRC catalytic but not scaffolding function is needed for integrin-regulated tyrosine phosphorylation, cell migration, and cell spreading. *Mol. Cell. Biol.* 22(8):2427-2440.
- 4. Eliceiri, B.P., et al. (2002) Src-mediated coupling of focal adhesion kinase to integrin alpha(v)beta5 in vascular endothelial growth factor signaling. *J. Cell Biol.* 157(1):149-160.
- Lim, Y., et al. (2002) Trichostatin A-induced detransformation correlates with decreased focal adhesion kinase phosphorylation at tyrosine 861 in ras-transformed fibroblasts. *J. Biol. Chem.* 277(15):12735-12740.
- 6. Cho, S.Y. and R.L. Klemke (2002) Purification of pseudopodia from polarized cells reveals redistribution and activation of Rac through assembly of a CAS/Crk scaffold. *J. Cell Biol.* 156(4):725-736.
- Rigacci, S., et al. (2002) Low Mr phosphotyrosine protein phosphatase associates and dephosphorylates p125 focal adhesion kinase, interfering with cell motility and spreading. *J. Biol. Chem.* 277(44):41631-41636.
- Kumar, P., et al. (2001) Soluble E-selectin induces monocyte chemotaxis through Src family tyrosine kinases. *J. Biol. Chem.* 276 (24):21039-21045.
- 9. Watcharasit, P., et al. (2001) Src family kinase involvement in muscarinic receptor-induced tyrosine phosphorylation in

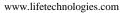
Explanation of symbols

Symbol	Description	Symbol	Description	Symbol	Description
***	Manufacturer	REF	Catalog number	LOT	Batch code
\square	Use by	X	Temperature limitation		
i	Consult instructions for use	\triangle	Caution, consult accompanying documents		

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References, continued

10. Sieg, D.J., et al. (2000) FAK integrates growth-factor and integrin signals to promote cell migration. *Nature Cell Biol.* 2(5):249-256.

Related products

Product Name	Quantity	Cat. No.
SRC [pY418] Polyclonal Antibody, Rabbit	10 blots	44660G
Pyk2 Antibody Sampler Pack	4 vials	44638G
FAK PSSA Antibody Sample Pack	5 vials	44631G

Product documentation

To obtain a Certificate of Analysis or Safety Data Sheets (SDSs), visit www.lifetechnologies.com/support.

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