SunLong Biotech Co.,LTD
Tel: 0086-571-56623320 Fax:0086-571-56623318
E-mail:sales@sunlongbiotech.com
www.sunlongbiotech.com

[Primary Antibody]

Rabbit Anti-Phospho-PAK4 (Ser99)antibody

Catalog Number: SL2270R

FOR RESEARCH USE ONLY

[DATASHEET]

Host:	Rabbit
Isotype:	IgG
Clonality:	Polyclonal
Target:	Phospho-PAK4 (Ser99)
Immunogen:	KLH conjugated Synthesised phosphopeptide derived from human PAK4 around the phosphorylation site of Ser99: SN(p-S)LR
Purification:	affinity purified by Protein A
Concentration:	1mg/ml
Storage:	0.01M TBS (pH7.4) with 1% BSA, 0.02% Proclin300 and 50% Glycerol. Shipped at 4℃. Store at -20 °C for one year. Avoid repeated freeze/thaw cycles.
Applications:	WB=1:500-2000 IHC-P=1:100-500 IHC-F=1:100-500 IF=1:100-500 Flow-Cyt=1ug/Test ELISA=1:5000-10000 （Paraffin sections need antigen repair） not yet tested in other applications. optimal dilutions/concentrations should be determined by the end user.
Reactivity:	Human,Mouse,Rabbit(predicted:Rat,Chicken,Dog,Cow,Horse)
Predicted MW.:	64kDa
Subcellular Location:	cytoplasmic
Background:	p21-activated kinases (PAKs) belong to the family of serine/threonine kinases involved in the control of various cellular processes, including the cell cycle, dynamics of the cytoskeleton, apoptosis, oncogenic transformation, and transcription. All PAK family members are characterized by the presence of p21-binding domain. p21-activated kinases are regulated by the small GTP-binding proteins Rac and Cdc42, and lipids, which stimulate autophosphorylation and phosphorylation of exogenous substrates. Serine (Ser-474) is the likely autophosphorylation site in the kinase domain of PAK4 in vivo. Phosphospecific antibodies directed against serine 474 detect activated

Important Note: This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.

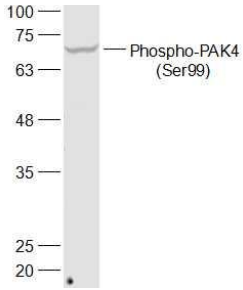
	PAK4 on the Golgi membrane when PAK4 is co-expressed with activated Cdc42. Current data strongly implicates PAK-4 in oncogenesis. PAK4 is frequently overexpressed in human tumor cell lines of various tissue origins. **Function:** Serine/threonine protein kinase that plays a role in a variety of different signaling pathways including cytoskeleton regulation, cell migration, growth, proliferation or cell survival. Activation by various effectors including growth factor receptors or active CDC42 and RAC1 results in a conformational change and a subsequent autophosphorylation on several serine and/or threonine residues. Phosphorylates and inactivates the protein phosphatase SSH1, leading to increased inhibitory phosphorylation of the actin binding/depolymerizing factor cofilin. Decreased cofilin activity may lead to stabilization of actin filaments. Phosphorylates LIMK1, a kinase that also inhibits the activity of cofilin. Phosphorylates integrin beta5/ITGB5 and thus regulates cell motility. Phosphorylates ARHGEF2 and activates the downstream target RHOA that plays a role in the regulation of assembly of focal adhesions and actin stress fibers. Stimulates cell survival by phosphorylating the BCL2 antagonist of cell death BAD. Alternatively, inhibits apoptosis by preventing caspase-8 binding to death domain receptors in a kinase independent manner. Plays a role in cell-cycle progression by controlling levels of the cell-cycle regulatory protein CDKN1A and by phosphorylating RAN. **Subunit:** Interacts with FGFR2 and GRB2. Interacts tightly with GTP-bound but not GDP-bound CDC42/p21 and weakly with RAC1. **Subcellular Location:** Cytoplasm. Note=Seems to shuttle between cytoplasmic compartments depending on the activating effector. For example, can be found on the cell periphery after activation of growth-factor or integrin-mediated signaling pathways. **Tissue Specificity:** Highest expression in prostate, testis and colon. **Post-translational modifications:** Autophosphorylated on serine residues when activated by CDC42/p21. Phosphorylated on tyrosine residues upon stimulation of FGFR2. **Similarity:** Belongs to the protein kinase superfamily. STE Ser/Thr protein kinase family. STE20 subfamily. Contains 1 CRIB domain. Contains 1 protein kinase domain. **Database links:** **Entrez Gene:** 10298 Human **Entrez Gene:** 70584 Mouse **Entrez Gene:** 292756 Rat Omim: 605451 Human

Important Note: This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.

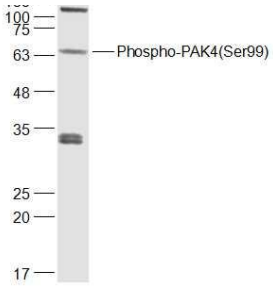
SunLong Biotech Co.,LTD
Tel: 0086-571-56623320 Fax:0086-571-56623318
E-mail:sales@sunlongbiotech.com
www.sunlongbiotech.com

	SwissProt: O96013 Human **SwissProt:** Q8BTW9 Mouse **Unigene:** 20447 Human **Unigene:** 21876 Mouse **Unigene:** 18543 Rat Serine/threonine-protein kinase 4 (PAK4) is a newly discovered class of protein factor that plays a very important role in cell Signal transduction. Pak4 is often overexpressed in Tumour cells of various human tissues and organs. Its serine site is very likely an autophosphorylation site in the enzymatic activity region of Pak4, and the serine-to-glutamate mutant confers enzymatic activity. It is of great significance in some important Cell biology studies such as Tumour genetics, cellular carcinogenesis, and Apoptosis.

[VALIDATION IMAGES]



Sample:
Hela(Human) Cell Lysate at 30 ug
Primary: Anti-Phospho-PAK4(Ser99) (SL2270R) at 1/500 dilution
Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution
Predicted band size: 64 kD
Observed band size: 64 kD



Sample:
293T(Human) Cell Lysate at 30 ug
Primary: Anti-Phospho-PAK4(Ser99) (SL2270R) at 1/1000 dilution
Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution
Predicted band size: 64 kD
Observed band size: 64 kD

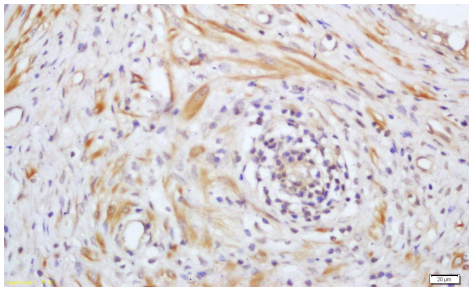
Important Note: This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.

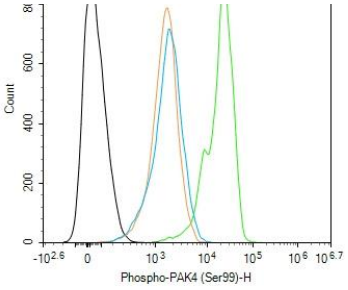
SunLong Biotech Co.,LTD

Tel: 0086-571-56623320 Fax:0086-571-56623318

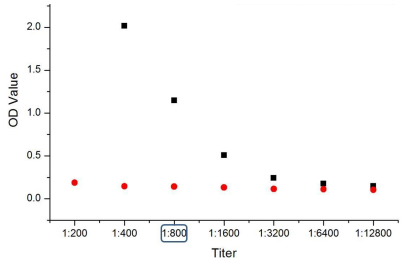
E-mail:sales@sunlongbiotech.com

www.sunlongbiotech.com





Tissue/cell: human prostate tissue; 4% Paraformaldehyde-fixed and paraffin-embedded;
Antigen retrieval: citrate buffer (Human,Mouse,Rabbit(predicted:Rat,Chicken,Dog,Cow,Horse)1M, pH 6.0), Boiling bathing for 15min; Block endogenous peroxidase by 3% Hydrogen peroxide for 30min; Blocking buffer (normal goat serum,C-0005) at 37℃ for 20 min;
Incubation: Anti-Phospho-PAK4 (Ser99) Polyclonal Antibody, Unconjugated(SL2270R) 1:200, overnight at 4°C, followed by conjugation to the secondary antibody(SP-0023) and DAB(C-0010) staining



phosphopeptide non phosphopeptide

Blank control（black line）:NIH/3T3. Primary Antibody (green line): Rabbit Anti-Phospho-PAK4 (Ser99) antibody (SL2270R) Dilution:1ug/Test; Secondary Antibody（white blue line）: Goat anti-rabbit IgG-AF488 Dilution: 0.5ug/Test. Isotype control（orange line）: Normal Rabbit IgG
Protocol The cells were fixed with 4% PFA (10min at room temperature)and then permeabilized with 90% ice-cold methanol for 20 min at -20℃, The cells were then incubated in 5%BSA to block non-specific protein-protein interactions for 30 min at room temperature .Cells stained with Primary Antibody for 30 min at room temperature. The secondary antibody used for 40 min at room temperature. Acquisition of 20,000 events was performed.

Important Note: This product as supplied is intended for research use only, not for use in human, therapeutic or diagnostic applications.