

**bs-2270R****[ Primary Antibody ]****phospho-PAK4 (Ser99) Rabbit pAb****Bioss**  
**ANTIBODIES**

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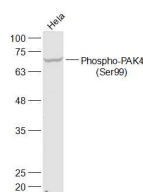
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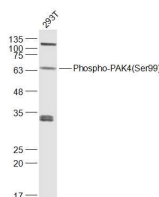
400-901-9800

**DATASHEET****Host:** Rabbit**Isotype:** IgG**Clonality:** Polyclonal**GeneID:** 10298**SWISS:** O96013**Target:** 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.

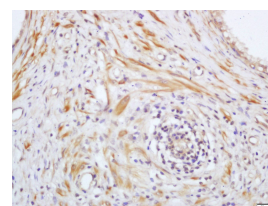
Shipped at 4°C. Store at -20°C for one year. Avoid repeated freeze/thaw cycles.

**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 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.**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)**Reactivity:** Human, Mouse, Rabbit  
(predicted: Rat, Cow, Chicken, Dog, Horse)**Predicted MW.:** 64 kDa**Subcellular Location:** Cytoplasm**VALIDATION IMAGES**

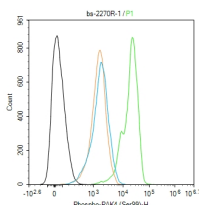
Sample: HeLa(Human) Cell Lysate at 30 ug  
 Primary: Anti-Phospho-PAK4(Ser99) (bs-2270R)  
 at 1/500 dilution Secondary: IRDye800CW Goat  
 Anti-Rabbit IgG at 1/20000 dilution Predicted  
 band size: 64 kD Observed band size: 64 kD



Sample: 293T(Human) Cell Lysate at 30 ug  
 Primary: Anti-Phospho-PAK4(Ser99) (bs-2270R)  
 at 1/1000 dilution Secondary: IRDye800CW Goat  
 Anti-Rabbit IgG at 1/20000 dilution Predicted  
 band size: 64 kD Observed band size: 64 kD



Tissue/cell: human prostate tissue; 4%  
 Paraformaldehyde-fixed and paraffin-  
 embedded; Antigen retrieval: citrate buffer (0.01M, 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°C for 20 min; Incubation:  
 Anti-Phospho-PAK4 (Ser99) Polyclonal Antibody,  
 Unconjugated(bs-2270R) 1:200, overnight at 4°C,  
 followed by conjugation to the secondary  
 antibody(SP-0023) and DAB(C-0010) staining



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

Blank control (black line) :NIH/3T3. Primary Antibody (green line): Rabbit Anti-Phospho-PAK4 (Ser99) antibody (bs-2270R) 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°C, 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.