Data Sheet (Cat.No.TMAB-11111)

TargetMol

Anti-Phospho-PAK6 (Ser560) Polyclonal Antibody

Product Details

Ig Type:	IgG
Reactivity:	Human,Mouse,Rat (predicted:Chicken,Dog,Pig,Cow,Horse,Rabbit,Sheep)
Molecular Weight:	Theoretical: 75 kDa. Actual: 73 kDa.
Purification:	Protein A purified

Applications

Verified Activity:

1. Paraformaldehyde-fixed, paraffin embedded (rat skin); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30 min; Antibody incubation with (Phospho-PAK6 (Ser560)) Polyclonal Antibody, Unconjugated (TMAB-11111) at 1:100 overnight at 4°C, followed by operating according to SP Kit (Rabbit) instructions and DAB staining.
2. Paraformaldehyde-fixed, paraffin embedded (rat brain); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30 min; Antibody incubation with (Phospho-PAK6 (Ser560)) Polyclonal Antibody, Unconjugated (TMAB-11111) at 1:100 overnight at 4°C, followed by operating according to SP Kit (Rabbit) instructions and DAB staining.
3. Paraformaldehyde-fixed, paraffin embedded (mouse brain); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30 min; Antibody incubation with (Phospho-PAK6 (Ser560)) Polyclonal Antibody, Unconjugated (TMAB-11111) at 1:100 overnight at 4°C, followed by operating according to SP Kit (Rabbit) instructions and DAB staining.
4. Paraformaldehyde-fixed, paraffin embedded (mouse skin); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30 min; Antibody incubation with (Phospho-PAK6 (Ser560)) Polyclonal Antibody, Unconjugated (TMAB-11111) at 1:100 overnight at 4°C, followed by operating according to SP Kit (Rabbit) instructions and DAB staining.
5. Paraformaldehyde-fixed, paraffin embedded (mouse esophagus); Antigen retrieval by boiling in sodium citrate buffer (pH6.0) for 15 min; Block endogenous peroxidase by 3% hydrogen peroxide for 20 minutes; Blocking buffer (normal goat serum) at 37°C for 30 min; Antibody incubation with (Phospho-PAK6 (Ser560)) Polyclonal Antibody, Unconjugated (TMAB-11111) at 1:100 overnight at 4°C, followed by operating according to SP Kit (Rabbit) instructions and DAB staining.
6. Sample:
Lane 1: Human PANC-1 cell lysates
Lane 2: Human A431 cell lysates
Lane 3: Human A549 cell lysates
Lane 4: Human MCF-7 cell lysates
Primary: Anti-Phospho-PAK6 (Ser560) (TMAB-11111) at 1/1000 dilution
Secondary: IRDye800CW Goat Anti-Rabbit IgG at 1/20000 dilution
Predicted band size: 75 kDa

Observed band size: 73 kDa
7. A431 cell; 4% Paraformaldehyde-fixed; Triton X-100 at room temperature for 20 min; Blocking buffer (normal goat serum) at 37°C for 20 min; Antibody incubation with (Phospho-PAK6 (Ser560)) polyclonal Antibody, Unconjugated (TMAB-11111) 1:100, 90 minutes at 37°C; followed by a conjugated Goat Anti-Rabbit IgG antibody at 37°C for 90 minutes, DAPI (blue) was used to stain the cell nuclei.
8. Blank control (black line): A549.
Primary Antibody (green line): Rabbit Anti-Phospho-PAK6 (Ser560) antibody (TMAB-11111)
Dilution: 1 μg/Test;
Secondary Antibody (white blue line): Goat anti-rabbit IgG-AF488
Dilution: 0.5 μg/Test.
Isotype control (orange line): Normal Rabbit IgG
Protocol
The cells were fixed with 4% PFA (10 min 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.

Application:	WB,IHC-P,IHC-Fr,ICC/IF,IF,FCM
Recommended	WB: 1:500-2000; IHC-P: 1:100-500; IHC-Fr: 1:100-500; ICC/IF: 1:100-500; IF: 1:100-500; FCM: 1μg/Test

Properties

Stability & Storage:	Store at 2°C-8°C for 1 month. Store at -20°C or -80°C for 12 months. Avoid repeated freeze-thaw cycles.
Shipping:	Shipping with blue ice.

Antigen Details

Immunogen:	KLH conjugated Synthesised phosphopeptide: human PAK6 around the phosphorylation site of Ser560
Antigen Species:	Human
Gene ID:	56924
Uniprot ID:	Q9NQU5

Research Background

This gene encodes a protein that shares a high degree of sequence similarity with p21-activated kinase (PAK) family members. The proteins of this family are Rac/Cdc42-associated Ste20-like Ser/Thr protein kinases, characterized by a highly conserved amino-terminal Cdc42/Rac interactive binding (CRIB) domain and a carboxyl-terminal kinase domain. PAK kinases are implicated in the regulation of a number of cellular processes, including cytoskeleton rearrangement, apoptosis and the MAP kinase signaling pathway. The protein encoded by this gene was found to interact with androgen receptor (AR), which is a steroid hormone-dependent transcription factor that is important for male sexual differentiation and development. The p21-activated protein kinase 6 gene was found to be highly expressed in testis and prostate tissues and the encoded protein was shown to cotranslocate into the nucleus with AR in response to androgen.

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