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Anti-PAK 1/2/3 pT423 (RABBIT) Antibody

PAK 1/2/3 phospho T423 Antibody
Catalog # ASR5207

Specification

Anti-PAK 1/2/3 pT423 (RABBIT) Antibody - Product Information

Host	**Rabbit**
Conjugate	**Unconjugated**
Target Species	**Human**
Reactivity	**Human**
Clonality	**Polyclonal**
Application	**WB, E, I, LCI**
Application Note	**This phospho specific polyclonal antibody was tested by Dot blot and ELISA and was found to be reactive with the phosphorylated form of the immunizing peptide and minimally reactive with the non-phosphorylated form of the immunizing peptide. Although not tested, this antibody is likely functional in immunohistochemistry, immunoblotting, and immunoprecipitation. Lysates from Jurkat cells or PAK transfected cells may be used as a control. This product has been assayed against 0.1 µg of phosphorylated peptide in a standard capture ELISA using TMB (3,3',5,5'-Tetramethylbenizidine) code # TMBE-100 as a substrate for 30 minutes at room temperature. A working dilution of 1:5,000 to 1:25,000 is suggested for this product. Less than 0.2% cross-reactivity was detected against the non-phosphorylated form of the immunizing peptide. Researchers should determine optimal titers for other applications.**
Physical State	**Liquid (sterile filtered)**
Buffer	**0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2**
Immunogen	**Human PAK 1/2/3 phospho peptide corresponding to a region of the human protein conjugated to Keyhole Limpet Hemocyanin (KLH).**
Preservative	**0.01% (w/v) Sodium Azide**

Anti-PAK 1/2/3 pT423 (RABBIT) Antibody - Additional Information

Gene ID 5058

Other Names
5058

Purity
This affinity purified antibody is directed against human PAK (p21-GTPase Activated Protein Kinase). The product was affinity purified from monospecific antiserum by immunoaffinity purification. Antiserum was first purified against the phosphorylated form of the immunizing peptide. The resultant affinity purified antibody was then cross-adsorbed against the non-phosphorylated form of the immunizing peptide. This phospho specific polyclonal antibody is specific for phosphorylated pT423 of Human PAK 1/2/3. Reactivity with non-phosphorylated PAK 1/2/3 is less than 1% by ELISA. Cross reactivity with PAK 1/2/3 from other species has not been determined.

Storage Condition
Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.

Precautions Note
This product is for research use only and is not intended for therapeutic or diagnostic applications.

Anti-PAK 1/2/3 pT423 (RABBIT) Antibody - Protein Information

Name PAK1 {ECO:0000303|PubMed:8805275, ECO:0000312|HGNC:HGNC:8590}

Function
Protein kinase involved in intracellular signaling pathways downstream of integrins and receptor-type kinases that plays an important role in cytoskeleton dynamics, in cell adhesion, migration, proliferation, apoptosis, mitosis, and in vesicle-mediated transport processes (PubMed:10551809, PubMed:11896197, PubMed:12876277, PubMed:14585966, PubMed:15611088, PubMed:17726028, PubMed:17989089, PubMed:30290153). Can directly phosphorylate BAD and protects cells against apoptosis (By similarity). Activated by interaction with CDC42 and RAC1 (PubMed:8805275, PubMed:9528787). Functions as a GTPase effector that links the Rho-related GTPases CDC42 and RAC1 to the JNK MAP kinase pathway (PubMed:8805275, PubMed:9528787). Phosphorylates and activates MAP2K1, and thereby mediates activation of downstream MAP kinases (By similarity). Involved in the reorganization of the actin cytoskeleton, actin stress fibers and of focal adhesion complexes (PubMed:9032240, PubMed:9395435). Phosphorylates the tubulin chaperone TBCB and thereby plays a role in the regulation of microtubule biogenesis and organization of the tubulin cytoskeleton (PubMed:15831477). Plays a role in the regulation of insulin secretion in response to elevated glucose levels (PubMed:22669945). Part of a ternary complex that contains PAK1, DVL1 and MUSK that is important for

MUSK-dependent regulation of AChR clustering during the formation of the neuromuscular junction (NMJ) (By similarity). Activity is inhibited in cells undergoing apoptosis, potentially due to binding of CDC2L1 and CDC2L2 (PubMed:12624090). Phosphorylates MYL9/MLC2 (By similarity). Phosphorylates RAF1 at 'Ser-338' and 'Ser-339' resulting in: activation of RAF1, stimulation of RAF1 translocation to mitochondria, phosphorylation of BAD by RAF1, and RAF1 binding to BCL2 (PubMed:11733498). Phosphorylates SNAI1 at 'Ser-246' promoting its transcriptional repressor activity by increasing its accumulation in the nucleus (PubMed:15833848). In podocytes, promotes NR3C2 nuclear localization (By similarity). Required for atypical chemokine receptor ACKR2-induced phosphorylation of LIMK1 and cofilin (CFL1) and for the up-regulation of ACKR2 from endosomal compartment to cell membrane, increasing its efficiency in chemokine uptake and degradation (PubMed:23633677). In synapses, seems to mediate the regulation of F- actin cluster formation performed by SHANK3, maybe through CFL1 phosphorylation and inactivation (By similarity). Plays a role in RUFY3-mediated facilitating gastric cancer cells migration and invasion (PubMed:25766321). In response to DNA damage, phosphorylates MORC2 which activates its ATPase activity and facilitates chromatin remodeling (PubMed:23260667). In neurons, plays a crucial role in regulating GABA(A) receptor synaptic stability and hence GABAergic inhibitory synaptic transmission through its role in F-actin stabilization (By similarity). In hippocampal neurons, necessary for the formation of dendritic spines and excitatory synapses; this function is dependent on kinase activity and may be exerted by the regulation of actomyosin contractility through the phosphorylation of myosin II regulatory light chain (MLC) (By similarity). Along with GIT1, positively regulates microtubule nucleation during interphase (PubMed:27012601). Phosphorylates FXR1, promoting its localization to stress granules and activity (PubMed:20417602).

Cellular Location
Cytoplasm. Cell junction, focal adhesion. Cell projection, lamellipodium. Cell membrane. Cell projection, ruffle membrane. Cell projection, invadopodium. Nucleus, nucleoplasm. Chromosome. Cytoplasm, cytoskeleton, microtubule organizing center, centrosome Note=Colocalizes with RUFY3, F-actin and other core migration components in invadopodia at the cell periphery (PubMed:25766321) Recruited to the cell membrane by interaction with CDC42 and RAC1 Recruited to focal adhesions upon activation. Colocalized with CIB1 within membrane ruffles during cell spreading upon readhesion to fibronectin. Upon DNA damage, translocates to the nucleoplasm when phosphorylated at Thr-212 where is co-recruited with MORC2 on damaged chromatin (PubMed:23260667). Localization to the centrosome does not depend upon the presence of gamma-tubulin (PubMed:27012601) Localization of the active, but not inactive, protein to the adhesions and edge of lamellipodia is mediated by interaction with GIT1 (PubMed:11896197). {ECO:0000250|UniProtKB:P35465, ECO:0000269|PubMed:11896197, ECO:0000269|PubMed:23260667, ECO:0000269|PubMed:25766321, ECO:0000269|PubMed:27012601}

Tissue Location
Overexpressed in gastric cancer cells and tissues (at protein level) (PubMed:25766321).

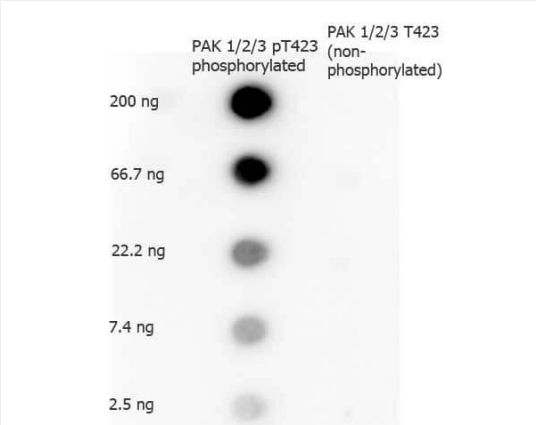
Anti-PAK 1/2/3 pT423 (RABBIT) Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

- Western Blot

- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

Anti-PAK 1/2/3 pT423 (RABBIT) Antibody - Images



Dot Blot of Rabbit anti-PAK 1/2/3 pT423 antibody. Antigen: phosphorylated and non-phosphorylated forms of the immunizing peptide. Load: 200 ng, 66.7 ng, 22.2 ng, 7.4 ng, or 2.5 ng as indicated. Primary antibody: PAK 1/2/3 pT423 antibody at 1:1,000 overnight at 4°C. Secondary antibody: Dylight™488 rabbit secondary antibody (p/n 611-141-002) at 1:40,000 for 45 min at RT. Block: Blocking Buffer for Fluorescent Western Blotting (MB-070) for 60 min at RT.

Anti-PAK 1/2/3 pT423 (RABBIT) Antibody - Background

The p21-activated kinases (PAKs) are a family of multifunctional serine/threonine kinases involved in a variety of cell functions including stress response, apoptosis and regulation of cell motility and tumor metastasis. Mammalian PAKs are called 1, 2, 3 or a, g, b respectively. PAKs are part of a large family of kinases where the catalytic domain of the kinase is related to Ste20 kinase of S. cerevisiae. Pak activity is regulated by specific binding of GTP-bound Rac and cdc42 GTPases and also by sphingosine and related lipids. PAK1 activation is induced by a variety of growth factors and G-protein-coupled receptors, Fc receptors, and integrins. This antibody is specific for the phosphorylated form of PAK 1/2/3. The selected peptide sequence used to generate the polyclonal antibody is common to all human PAKs.