

## Product datasheet

# Anti-PAK1+PAK2+PAK3 (phospho S141 + S144 + S154) antibody [EP656Y] - BSA and Azide free ab239830

Recombinant RabMAb

8 Images

### Overview

<b>Product name</b>	Anti-PAK1+PAK2+PAK3 (phospho S141 + S144 + S154) antibody [EP656Y] - BSA and Azide free
<b>Description</b>	Rabbit monoclonal [EP656Y] to PAK1+PAK2+PAK3 (phospho S141 + S144 + S154) - BSA and Azide free
<b>Host species</b>	Rabbit
<b>Tested applications</b>	<b>Suitable for:</b> Flow Cyt (Intra), IP, IHC-P, ICC/IF, WB
<b>Species reactivity</b>	<b>Reacts with:</b> Mouse, Rat, Human
<b>Immunogen</b>	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
<b>Positive control</b>	WB: MCF7, HeLa, RAW 264.7 and C6 cell lysates. IHC: Human liver carcinoma, mouse cerebral cortex, rat cerebral cortex. ICC/IF: HeLa cells. IP: HeLa cell lysate. Flow Cyt (intra): NIH/3T3 cell lysate.
<b>General notes</b>	<p>ab239830 is the carrier-free version of <a href="#">ab40795</a>.</p> <p>Our <b>carrier-free</b> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.</p> <p>This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.</p> <p>Use our <b>conjugation kits</b> for antibody conjugates that are ready-to-use in as little as 20 minutes with &lt;1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.</p> <p>This product is compatible with the Maxpar<sup>®</sup> Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar<sup>®</sup> is a trademark of Fluidigm Canada Inc.</p> <p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"><li>- High batch-to-batch consistency and reproducibility</li><li>- Improved sensitivity and specificity</li><li>- Long-term security of supply</li><li>- Animal-free production</li></ul> <p>For more information <a href="#">see here</a>.</p>

Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to [RabMAb<sup>®</sup> patents](#).

## Properties

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<b>Form</b>	Liquid
<b>Storage instructions</b>	Shipped at 4°C. Store at +4°C. Do Not Freeze.
<b>Storage buffer</b>	pH: 7.2 Constituent: PBS
<b>Carrier free</b>	Yes
<b>Purity</b>	Protein A purified
<b>Clonality</b>	Monoclonal
<b>Clone number</b>	EP656Y
<b>Isotype</b>	IgG

## Applications

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**The Abpromise guarantee** Our [Abpromise guarantee](#) covers the use of ab239830 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
<b>Flow Cyt (Intra)</b>		Use at an assay dependent concentration. <b>ab199376</b> - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
<b>IP</b>		Use at an assay dependent concentration.
<b>IHC-P</b>		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
<b>ICC/IF</b>		Use at an assay dependent concentration.
<b>WB</b>		Use at an assay dependent concentration. Detects a band of approximately 66 kDa (predicted molecular weight: 65 kDa).

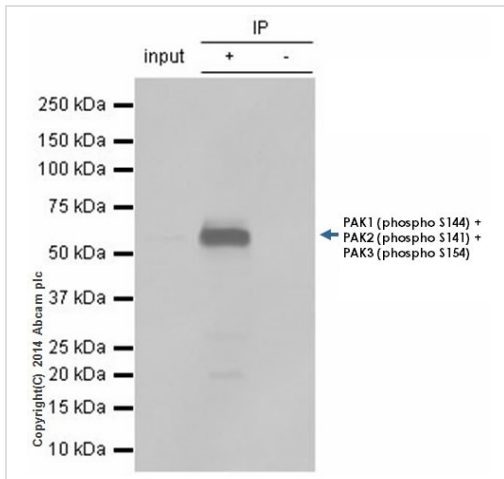
## Target

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**Cellular localization** PAK1: Cytoplasm. Cell junction > focal adhesion. Recruited to focal adhesions upon activation. PAK2: Cytoplasm and Nucleus. Cytoplasm > perinuclear region. Membrane. Interaction with ARHGAP10 probably changes PAK-2p34 location to cytoplasmic perinuclear region. Myristoylation changes PAK-2p34 location to the membrane. PAK3: Cytoplasmic

## Images

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Immunoprecipitation - Anti-PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) antibody [EP656Y] (ab239830)

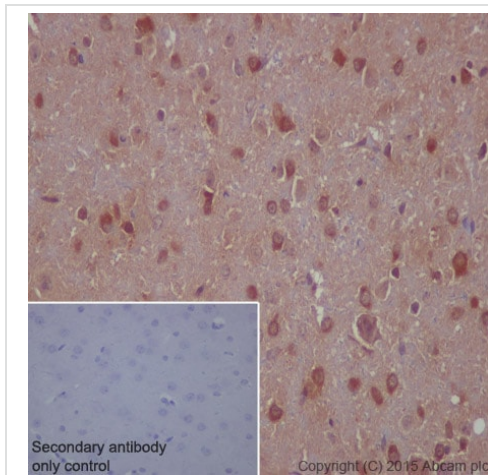
**ab40795** immunoprecipitating PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154). 10µg of HeLa (human cervix adenocarcinoma) whole cell lysate was incubated with primary antibody at a dilution of 1/40 and VeriBlot for IP Detection Reagent (HRP) (**ab131366**) at a dilution of 1/10000.

**Lane 1:** HeLa whole cell lysate (10ug)

**Lane 2:** **ab40795** IP in HeLa whole cell lysate

**Lane 3:** Rabbit monoclonal IgG (**ab172730**) instead of **ab40795** in HeLa (human cervix adenocarcinoma) whole cell lysate

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40795**).

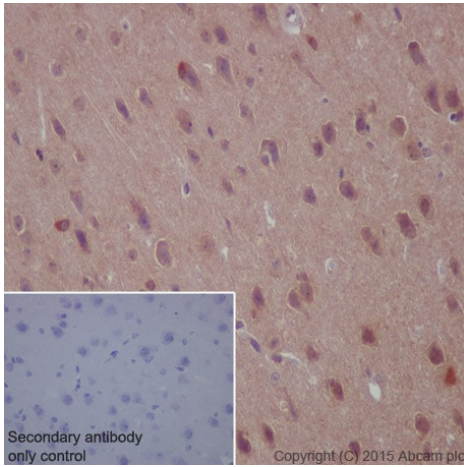


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) antibody [EP656Y] (ab239830)

**ab40795** staining PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) in rat cerebral cortex tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with paraformaldehyde and antigen retrieval was by heat mediation in a EDTA buffer. Samples were incubated with primary antibody at a dilution of 1/100. A goat anti-rabbit IgG H&L (HRP) **ab97051** was used as the secondary antibody at a dilution of 1/500.

**Negative control 1:** PBS in place of primary

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40795**).

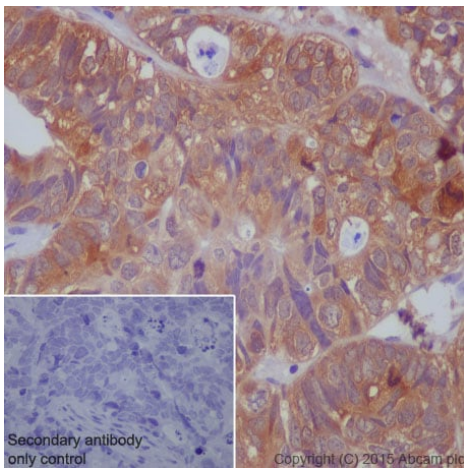


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) antibody [EP656Y] (ab239830)

**ab40795** staining PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) in mouse cerebral cortex tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with paraformaldehyde and antigen retrieval was by heat mediation in a EDTA buffer. Samples were incubated with primary antibody at a dilution of 1/100. A goat anti-rabbit IgG H&L (HRP) **ab97051** was used as the secondary antibody at a dilution of 1/500.

**Negative control 1:** PBS in place of primary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40795**).

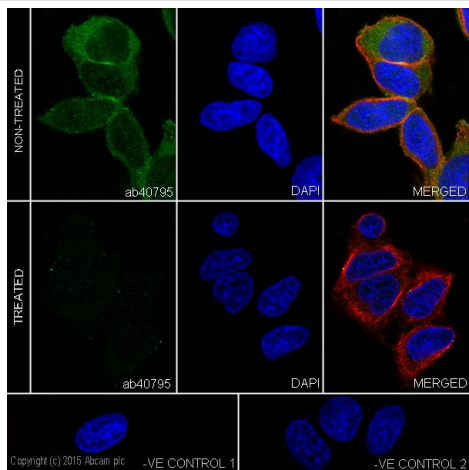


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) antibody [EP656Y] (ab239830)

**ab40795** staining PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) in human liver carcinoma tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with paraformaldehyde and antigen retrieval was by heat mediation in a EDTA buffer. Samples were incubated with primary antibody at a dilution of 1/100. A goat anti-rabbit IgG H&L (HRP) **ab97051** was used as the secondary antibody at a dilution of 1/500.

**Negative control 1:** PBS in place of primary antibody.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40795**).



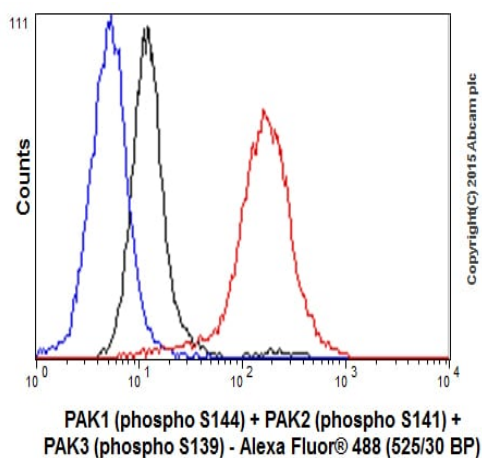
Immunocytochemistry/ Immunofluorescence - Anti-PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) antibody [EP656Y] (ab239830)

**ab40795** staining PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) in HeLa (human cervix adenocarcinoma) cells, treated and untreated with Lambda Protein Phosphatase 31? for 5h by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. Samples were incubated with primary antibody at a dilution of 1/100. A goat anti rabbit IgG (Alexa Fluor® 488) (**ab150077**) was used as the secondary antibody. **ab7291** and **ab150120** were used as counterstains for primary antibody **ab75748** and secondary antibody **ab150077** respectively and DAPI was used as a nuclear counterstain.

**Negative control 1:** Rabbit primary antibody and anti-mouse secondary antibody (**ab150120**)

**Negative control 2:** Mouse primary antibody (**ab7291**) and anti-rabbit secondary antibody (**ab150077**)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40795**).



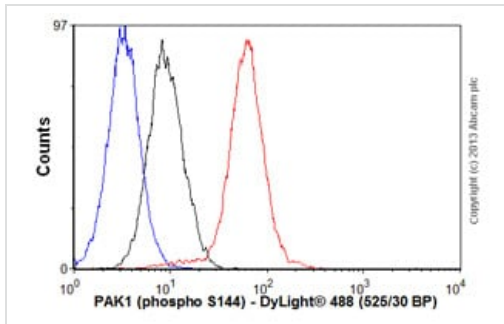
Flow Cytometry (Intracellular) - Anti-PAK1+PAK2+PAK3 (phospho S141 + S144 + S154) antibody [EP656Y] - BSA and Azide free (ab239830)

**ab40795** staining PAK1 (phospho S144) + PAK2 (phospho S141) + PAK3 (phospho S154) in the mouse cell line NIH/3T3 (mouse embryo) by intracellular flow cytometry. Cells were fixed with 4% paraformaldehyde and the sample was incubated with the primary antibody at a dilution of 1/120. A goat anti rabbit IgG (Alexa Fluor® 488) at a dilution of 1/500 was used as the secondary antibody.

Isotype control: Rabbit monoclonal IgG (Black)

Unlabelled control: Cell without incubation with primary antibody and secondary antibody (Blue)

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40795**).



Flow Cytometry (Intracellular) - Anti-PAK1+PAK2+PAK3 (phospho S141 + S144 + S154) antibody [EP656Y] - BSA and Azide free (ab239830)

Overlay histogram showing HeLa (Human epithelial cell line from cervix adenocarcinoma) cells stained with unpurified **ab40795** (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (**ab40795**, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (**ab96899**) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10<sup>6</sup> cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab40795**).

Why choose a recombinant antibody?

 <b>Research with confidence</b> Consistent and reproducible results	 <b>Long-term and scalable supply</b> Recombinant technology
 <b>Success from the first experiment</b> Confirmed specificity	 <b>Ethical standards compliant</b> Animal-free production

Anti-PAK1+PAK2+PAK3 (phospho S141 + S144 + S154) antibody [EP656Y] - BSA and Azide free (ab239830)

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