# abcam

# Product datasheet

# Anti-PYK2 antibody [YE353] ab32571





29 References 12 Images

Overview

**Product name** Anti-PYK2 antibody [YE353]

Rabbit monoclonal [YE353] to PYK2 **Description** 

**Host species** Rabbit

**Specificity** This antibody recognizes PYK2. It does not cross react with other FAK family members.

**Tested applications** Suitable for: WB, IHC-P, ICC/IF

Unsuitable for: **P** 

Reacts with: Mouse, Rat, Human Species reactivity

Synthetic peptide within Human PYK2 aa 1-100 (N terminal). The exact sequence is proprietary. **Immunogen** 

Database link: Q14289

Positive control WB: Ramos, Jurkat and RAW264.7 cell lysates and mouse and rat brain tissue lysates. IHC-P:

Human, mouse and rat cerebral cortex tissues. ICC/IF: HeLa and PC12 cells.

General notes This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

**Properties** 

**Form** Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C.

Avoid freeze / thaw cycle.

pH: 7.20 Storage buffer

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol, 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal

Clone number YE353

**Isotype** IgG

#### **Applications**

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab32571 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
WB		1/2000. Detects a band of approximately 116 kDa (predicted molecular weight: 116 kDa).  For unpurified use at 1/1000 - 1/5000.
IHC-P		1/300. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.  For unpurified use at 1/250 - 1/500.
ICC/IF		1/60. For unpurified use at 1/100.

**Application notes** 

Is unsuitable for IP.

#### **Target**

**Function** 

Involved in calcium induced regulation of ion channel and activation of the map kinase signaling pathway. May represent an important signaling intermediate between neuropeptide activated receptors or neurotransmitters that increase calcium flux and the downstream signals that regulate neuronal activity. Interacts with the SH2 domain of Grb2. May phosphorylate the voltage-gated potassium channel protein Kv1.2. Its activation is highly correlated with the stimulation of c-Jun N-terminal kinase activity. Involved in osmotic stress-dependent SNCA 'Tyr-125' phosphorylation. In concert with SRC, plays an important role in osteoclastic bone resorption. Both the formation of a SRC-PTK2B complex, and SRC kinase activity are necessary for this function. The Tyr-402 phosphorylated form serves as a docking site for SRC and is important for the organization of the osteoclast actin cytoskeleton and attachment sites and for bone resorption.

Tissue specificity

Most abundant in the brain, with highest levels in amygdala and hippocampus. Low levels in kidney. Also expressed in spleen and lymphocytes.

Sequence similarities

Belongs to the protein kinase superfamily. Tyr protein kinase family. FAK subfamily.

Contains 1 FERM domain.

Contains 1 protein kinase domain.

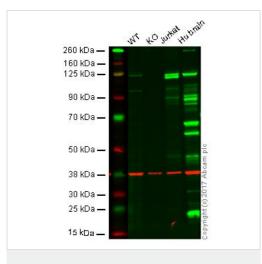
Post-translational modifications

Phosphorylated on tyrosine residues in response to various stimuli that elevate the intracellular calcium concentration, as well as by PKC activation. Recruitment by nephrocystin to cell matrix adhesions initiates Tyr-402 phosphorylation. In monocytes, adherence to substrata is required for tyrosine phosphorylation and kinase activation. Angiotensin II, thapsigargin and L-alphalysophosphatidic acid (LPA) also induce autophosphorylation and increase kinase activity.

**Cellular localization** 

Cytoplasm. Cell membrane. Interaction with nephrocystin induces the membrane-association of

the kinase.



Western blot - Anti-PYK2 antibody [YE353] (ab32571)



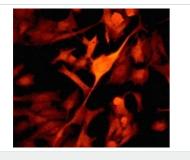
Lane 2: PYK2 knockout HAP1 whole cell lysate (20 µg)

Lane 3: Jurkat whole cell lysate (20 µg)

Lane 4: Hu brain whole cell lysate (20 µg)

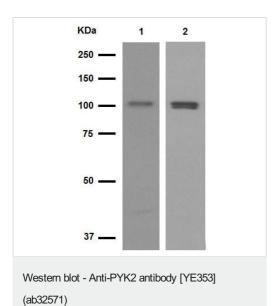
**Lanes 1 - 4:** Merged signal (red and green). Green - ab32571 observed at 125 kDa. Red - loading control, **ab8245**, observed at 37 kDa.

ab32571 was shown to specifically recognize PYK2 in wild-type HAP1 cells along with additional cross reactive bands. No band was observed when PYK2 knockout samples were examined. Wild-type and PYK2 knockout samples were subjected to SDS-PAGE. Ab32571 and <a href="mailto:ab8245">ab8245</a> (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed <a href="mailto:ab216773">ab216773</a> and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed <a href="mailto:ab216776">ab216776</a> secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-PYK2 antibody [YE353] (ab32571)

Immunocytochemistry/Immunofluorescence analysis of PC12 cells labelling PYK2 with unpurified ab32571 at a 1/100 dilution.



All lanes: Anti-PYK2 antibody [YE353] (ab32571) at 1/10000 dilution (purified)

Lane 1: Ramos cell lysate Lane 2: Jurkat cell lysate

Lysates/proteins at 20 µg per lane.

### Secondary

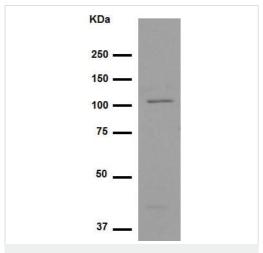
All lanes: Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Predicted band size: 116 kDa

Observed band size: 116 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-PYK2 antibody [YE353]

(ab32571)

Anti-PYK2 antibody [YE353] (ab32571) at 10000 cells (purified) + RAW264.7 cell lysate at 20 µg

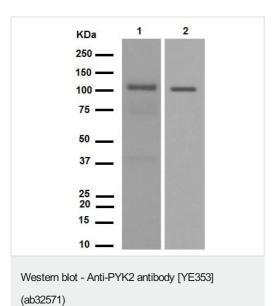
#### Secondary

Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

Predicted band size: 116 kDa Observed band size: 116 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



All lanes: Anti-PYK2 antibody [YE353] (ab32571) at 1/2000 dilution (purified)

Lane 1: Mouse brain tissue lysate

Lane 2: Rat brain tissue lysate

Lysates/proteins at 20 µg per lane.

## Secondary

All lanes: Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Predicted band size: 116 kDa

Observed band size: 116 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

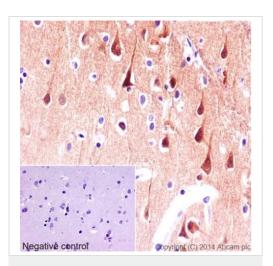
Diluting buffer and concentration: 5% NFDM /TBST.

250-150-50-37-

Western blot - Anti-PYK2 antibody [YE353] (ab32571)

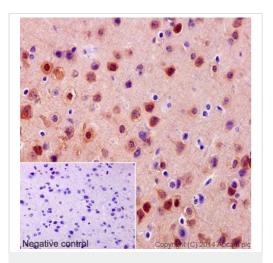
Anti-PYK2 antibody [YE353] (ab32571) at 1/5000 dilution (unpurified) + Jurkat cell lysate

Predicted band size: 116 kDa Observed band size: 116 kDa



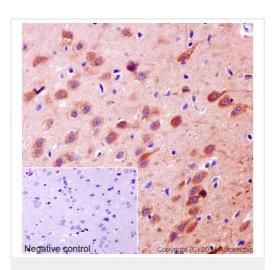
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PYK2 antibody [YE353] (ab32571)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human cerebral cortex tissue labelling PYK2 with purified ab32571 at 1/300. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. <a href="mailto:ab97051">ab97051</a>, a HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



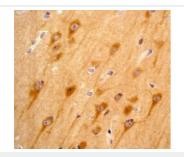
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PYK2 antibody [YE353] (ab32571)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse cerebral cortex tissue labelling PYK2 with purified ab32571 at 1/300. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. <a href="mailto:ab97051">ab97051</a>, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



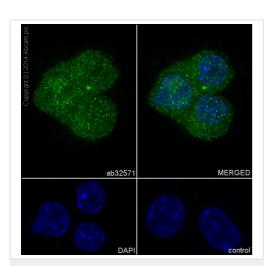
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PYK2 antibody [YE353] (ab32571)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat cerebral cortex tissue labelling PYK2 with purified ab32571 at 1/300. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. <a href="mailto:ab97051">ab97051</a>, a HRP-conjugated goat anti-rabbit lgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PYK2 antibody [YE353] (ab32571)

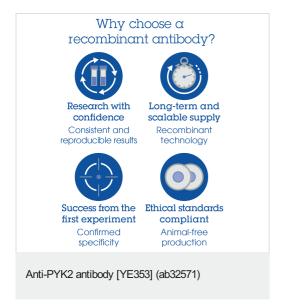
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of normal human brain tissue labelling PYK2 with unpurified ab32571 at a 1/250 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-PYK2 antibody [YE353] (ab32571)

Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling PYK2 with purified ab32571 at 1/60. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

Control: primary antibody (1/100) and secondary antibody, **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).



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