# abcam

# Product datasheet

# Anti-PCK2 antibody ab70359

# ★★★★★ 1 Abreviews 17 References 4 Images

#### Overview

Product name Anti-PCK2 antibody

**Description** Rabbit polyclonal to PCK2

Host species Rabbit

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

Species reactivity Reacts with: Human

Predicted to work with: Mouse, Rat, Pig

**Immunogen** Synthetic peptide corresponding to Human PCK2 aa 400-500 conjugated to keyhole limpet

haemocyanin.

(Peptide available as ab87120)

**Positive control** This antibody gave a positive signal in HepG2 Whole Cell Lysate.

**General notes**The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

#### **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 or -

80°C. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

scientific support team who will be happy to help.

**Purity** Immunogen affinity purified

1

**Clonality** Polyclonal

**Isotype** IgG

#### **Applications**

# The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab70359 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
ICC/IF		Use a concentration of 1 µg/ml.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
WB	**** (1)	Use a concentration of 1 µg/ml. Detects a band of approximately 71 kDa (predicted molecular weight: 71 kDa).
IP		Use at an assay dependent concentration.

Function Catalyzes the conversion of oxaloacetate (OAA) to phosphoenolpyruvate (PEP), the rate-limiting

step in the metabolic pathway that produces glucose from lactate and other precursors derived

from the citric acid cycle.

**Pathway** Carbohydrate biosynthesis; gluconeogenesis.

Involvement in disease Defects in PCK2 are the cause of mitochondrial phosphoenolpyruvate carboxykinase deficiency

(M-PEPCKD) [MIM:261650]. A metabolic disorder resulting from impaired gluconeogenesis. It is a rare disease with less than 10 cases reported in the literature. Clinical characteristics include hypotonia, hepatomegaly, failure to thrive, lactic acidosis and hypoglycemia. Autoposy reveals

 $fatty\ infiltration\ of\ both\ the\ liver\ and\ kidneys.\ The\ disorder\ is\ transmitted\ as\ an\ autosomal$ 

recessive trait.

Sequence similarities Belongs to the phosphoenolpyruvate carboxykinase [GTP] family.

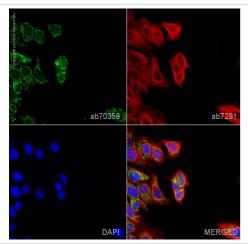
Post-translational

modifications

Phosphorylated upon DNA damage, probably by ATM or ATR.

Cellular localization Mitochondrion.

## **Images**



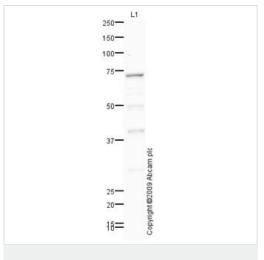
Immunocytochemistry/ Immunofluorescence - Anti-PCK2 antibody (ab70359)

Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.

DNA was labelled with DAPI (shown in blue).

ab70359 staining Phosphoenolpyruvate carboxykinase [GTP], mitochondrial precursor in MCF7 cells. The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.1% PBS-Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then

incubated overnight at 4°C with ab70359 at 1µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and ab150120, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), preadsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear



Western blot - Anti-PCK2 antibody (ab70359)

Anti-PCK2 antibody (ab70359) at 1 µg/ml + HepG2 (Human hepatocellular liver carcinoma cell line) Whole Cell Lysate at 10 µg

#### **Secondary**

Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

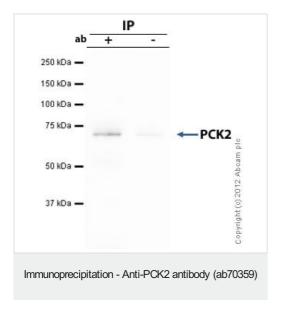
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 71 kDa Observed band size: 71 kDa

Additional bands at: 40 kDa, 50 kDa. We are unsure as to the

identity of these extra bands.

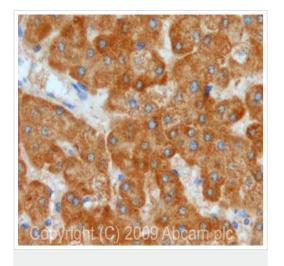


PCK2 was immunoprecipitated using 0.5mg HepG2 whole cell extract, 5µg of Rabbit polyclonal to PCK2 and 50µl of protein G magnetic beads (+). No antibody was added to the control (-). The antibody was incubated under agitation with Protein G beads for 10min, HepG2 whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab70359.

Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).

Band: 71kDa: PCK2.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PCK2 antibody (ab70359)

IHC image of PCK2 staining in normal human liver formalin fixed paraffin embedded tissue section, performed on a Leica Bond TM system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with <a href="mailto:ab70659">ab70659</a>, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

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