

Phospho-PKM2 (Tyr105) Antibody



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For Research Use Only. Not For Use In Diagnostic Procedures.

Applications	Species Cross-Reactivity*	Molecular Wt.	Source
W Endogenous	H, M, R, Mk	60 kDa	Rabbit**

Background: Pyruvate kinase is a glycolytic enzyme that catalyses the conversion of phosphoenolpyruvate to pyruvate. In mammals, the M1 isoform (PKM1) is expressed in most adult tissues (1). The M2 isoform (PKM2) is an alternatively spliced variant of M1 that is expressed during embryonic development (1). Research studies found that cancer cells exclusively express PKM2 (1-3). PKM2 is shown to be essential for aerobic glycolysis in tumors, known as the Warburg effect (1). When cancer cells switch from the M2 isoform to the M1 isoform, aerobic glycolysis is reduced and oxidative phosphorylation is increased (1). These cells also show decreased tumorigenicity in mouse xenografts (1). Recent studies showed that PKM2 is not essential for all tumor cells (4). In the tumor model studied, PKM2 was found to be active in the non-proliferative tumor cell population and inactive in the proliferative tumor cell population (4).

Additional studies show that the oncogenic forms of FGFR1 directly phosphorylate Tyr105 of PKM2 and thereby inhibit the formation of active, tetrameric PKM2 (5). A PKM2 mis-sense mutation found in cancer cells results in the replacement of Tyr105 by phenylalanine and leads to reduced cell proliferation during hypoxia and tumor growth in nude mice xenografts (5). These findings suggest that the phosphorylation at Tyr105 is a critical switch for the metabolism in cancer cells that promotes tumor growth (5).

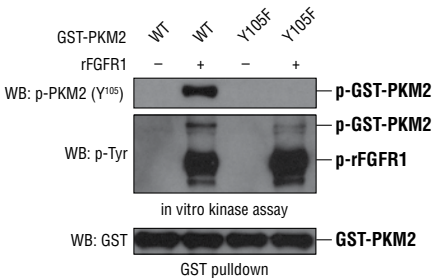
Phosphorylation of PKM2 on Tyr105 was identified at Cell Signaling Technology (CST) using PhosphoScan®, CST's LC-MS/MS platform for phosphorylation site discovery.

Specificity/Sensitivity: Phospho-PKM2 (Tyr105) Antibody detects endogenous levels of PKM2 protein only when phosphorylated at Tyr105. This antibody may slightly cross react with PKM1 phosphorylated at the equivalent site.

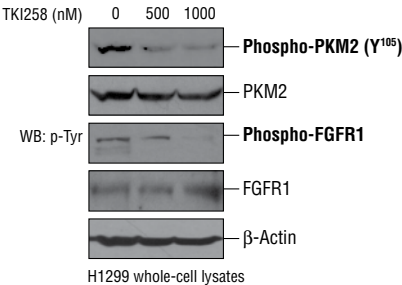
Source/Purification: Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to the sequence surrounding Tyr105 of human PKM2 protein. Antibodies are purified by protein A and peptide affinity chromatography.

Background References:

- (1) Christofk, H.R. et al. (2008) *Nature* 452, 230-3.
- (2) Mazurek, S. et al. (2005) *Semin Cancer Biol* 15, 300-8.



GST-PKM2 wild-type or the Tyr105Phe mutant was incubated in an in vitro kinase assay in the presence or absence of active FGFR1. Western blot analysis was performed using Phospho-PKM2 (Tyr105) Antibody and a phospho-Tyr antibody. The data demonstrate the specificity of the Phospho-PKM2 (Tyr105) Antibody and that the Tyr105Phe mutation abolishes PKM2 phosphorylation at Tyr105 by FGFR1 in vitro. (Adapted from Hitosugi, T. et al., 2009).



Western blot analysis of NCI-H1299 cells using Phospho-PKM2 (Tyr105) Antibody, total PKM2 Antibody #3198, total FGFR1 antibody, phospho-Tyr antibody, and β -actin antibody. The data demonstrate that inhibition of FGFR1 by TKI258 treatment in NCI-H1299 cells results in decreased Tyr105 phosphorylation of endogenous PKM2. (Adapted from Hitosugi, T. et al., 2009).

- (3) Dombravckas, J.D. et al. (2005) *Biochemistry* 44, 9417-29.
- (4) Israelsen, W.J. et al. (2013) *Cell* 155, 397-409.
- (5) Hitosugi, T. et al. (2009) *Sci Signal* 2, ra73.

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween® 20 at 4°C with gentle shaking, overnight.

Entrez-Gene ID #5315

Swiss-Prot Acc. #P14618

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 μ g/ml BSA and 50% glycerol. Store at -20°C. Do not aliquot the antibody.

*Species cross-reactivity is determined by western blot.

**Anti-rabbit secondary antibodies must be used to detect this antibody.

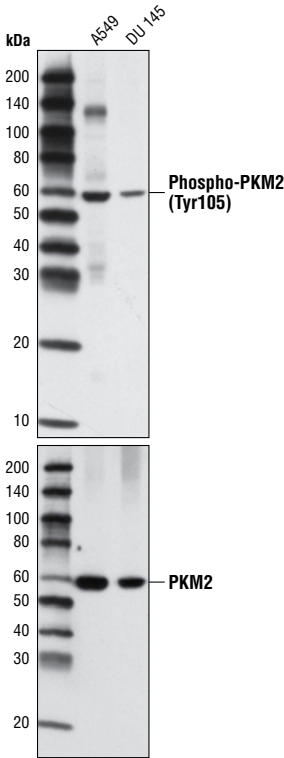
Recommended Antibody Dilutions:

Western blotting

1:1000

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.



Western blot analysis of extracts from A549 and DU 145 cells using Phospho-PKM2 (Tyr105) Antibody (upper) or PKM2 Antibody #3198 (lower).

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Applications Key:

W—Western

IP—Immunoprecipitation

IHC—Immunohistochemistry

ChIP—Chromatin Immunoprecipitation

IF—Immunofluorescence

F—Flow cytometry

E-P—ELISA-Peptide

Species Cross-Reactivity Key:

H—human

M—mouse

R—rat

Hm—hamster

Mk—monkey

Mi—mink

C—chicken

Dm—D. melanogaster

X—Xenopus

Z—zebrafish

B—bovine

Dg—dog

Pg—pig

Sc—S. cerevisiae

Ce—C. elegans

Hr—horse

All—all species expected

Species enclosed in parentheses are predicted to react based on 100% homology.