ALPHA DIAGNOSTIC INTERNATIONAL

Product Specification Sheet

Pyruvate Kinase L/R (PKLR; PK1; PKL**) Antibodies**

Cat # PKRL13-P	Human PKLR Control/Blocking Peptide	**SIZE**: 100 µg
Cat # PKRL13-A	Rabbit anti-human PKLR IgG (affinity pure)	**SIZE**: 100 µg

Glycolysis occurs in the cytosol, yielding 2 ATP, 2 pyruvate, and 2 NADH + 2 H+ from each glucose molecule. The first two steps of glycolysis convert glucose-6-phosphate into a form that is easily cleaved into phosphorylated three-carbon units. High energy phosphate (as ATP) is generated from the three-carbon units in the remaining steps. The absence of glucose-6-phosphatase from most tissues (exceptions are liver and kidney) makes glucose uptake by these tissues essentially irreversible, again consistent with the view that glucose is taken up for local metabolic use.

Pyruvate kinase is an enzyme involved in glycolysis. It catalyzes the transfer of a phosphoryl group from phosphoenolpyruvate to ADP, yielding a pyruvate molecule. There are 4 isozymes of pyruvate kinase in mammals: L, R, M1 and M2. L type is major isozyme in the liver, R is found in red cells, M1 is the main form in muscle, heart and brain, and M2 is found in early fetal tissues. The L and R isozymes are generated from the PKLR gene by differential splicing of RNA; the M1 and M2 forms are produced from the PKM2 gene by differential splicing.

PKLR: The protein encoded by this gene is a pyruvate kinase that catalyzes the production of phohsphoenolpyruvate from pyruvate and ATP. Defects in this enzyme, due to gene mutations or genetic variations, are the common cause of chronic hereditary nonspherocytic hemolytic anemia (CNSHA or HNSHA). This autosomal dominant phenotype is characterized by increase of red blood cell ATP. Alternatively spliced transcript variants encoding distinct isoforms have been described. Strong genetic evidences show that the L- and R-type PK enzymes are encoded by the same structural gene. Both in vitro and in vivo studies have shown that the rat R- and L-specific PK enzymes are produced from different transcription units operating with 2 cell-restricted promoters which, due to protein-DNA interactions, are mutually exclusive.

PKLR: human, rat, mouse: 574 aa each – 61.8 kDa; human chromosome: 1q21. Highly expressed in liver and RBCs.

SIMILARITY: Belongs to the pyruvate kinase family.
Protein name Pyruvate kinase L
Synonym EC 2.7.1.40

Source of Antigen and Antibodies

Antigen	17-aa peptide of human PKRL13-P, (protein accession #O75758 , refs 1) **designated as control peptide.** conjugated to KLH; Epitope location ~ N-terminal
Ab Host/type	Rabbit, polyclonal Aff pure IgG (cat#PKRL-A) purified over the antigen column
2-Ab	Anti-rabbit IgG-HRP cat # 20320 (AP, biotin, FITC conjugates also available)
-ve control	# 20009-1, Rabbit (non-immune) IgG, purified, suitable for ELISA, Western, IHC as –ve control

Form & Storage of Antibodies/Peptide Control

Affinity pure IgG
100 ug/100ul solution lyophilized powder
Supplied in **Buffer**: PBS+0.1% BSA
Reconstitute powder in PBS at 1mg/ml

Control/blocking peptide
100 ug/100 ul solution lyophilized powder
Supplied in Buffer: PBS pH 7.5,
Reconstitute powder in PBS at 1 mg/ml.

Storage

Short-term: unopened, undiluted liquid vials at -20^{O}C and powder at 4^{o}C or -20^{o}C..

Long-term: at –20^{0}C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at –20^{o}C or below.
Shipping: 4^{o}C for solutions and room temp for powder

Recommended Usage

Western Blotting: 1-10 µg/ml; using affinity pure antibody (chemiluminescence technique).

ELISA: 1:100K; using 50-100 ng control peptide/well.

Histochemistry & Immunofluorescence: Not tested; we recommend the use of affinity purified antibody at 2-10 µg/ml.

Specificity & Cross-reactivity

Human PKRL13-P peptide sequence is 93%, 87% and 81% conserved in mouse, rat and bovine respectively. Antibody cross-reactivity in various species is not known. The control peptide, because of its low mol. Wt (<3 kDa), is not suitable for Western. It should be used for ELISA or antibody blocking experiments (use 5-30 ug control peptide per 1 ug of aff pure IgG or 1 ul antiserum) to confirm antibody specificity

General References:

1. Tani K., Fujii H., Nagata S., Miwa S.; Proc. Natl. Acad. Sci. U.S.A. 85:1792-1795(1988).
2. Kanno H., Fujii H., Tsujino G., Miwa S.; Biochem. Biophys. Res. Commun. 192:46-52 (1993).
3. Beutler E., Baronciani L.; Hum. Mutat. 7:1-6(1996).

*This product is for in vitro research use only.

Related material available from ADI

PKR11-P; PKR11-A; PKR12P; PKR12-A
PKM14-P; PKM14-A

KRL13-A-P 70912J

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