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PKM2 contributes to cancer metabolism

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ABSTRACT

Reprogramming of cell metabolism is essential for tumorigenesis, and is regulated by a complex network, in which PKM2 plays a critical role. PKM2 exists as an inactive monomer, less active dimer and active tetramer. While dimeric PKM2 diverts glucose metabolism towards anabolism through aerobic glycolysis, tetrameric PKM2 promotes the flux of glucose-derived carbons for ATP production via oxidative phosphorylation. Equilibrium of the PKM2 dimers and tetramers is critical for tumorigenesis, and is controlled by multiple factors. The PKM2 dimer also promotes aerobic glycolysis by modulating transcriptional regulation. We will discuss the current understanding of PKM2 in regulating cancer metabolism.

1. Introduction

Cancer cells elevate glucose uptake with a concomitant increase of lactate production in a well-oxygenized environment; a process known as aerobic glycolysis or the Warburg effect [1,2]. While non-proliferating cells maximize ATP production under normoxic conditions through oxidative phosphorylation of glucose, proliferating cells utilize glucose both for ATP production and for the synthesis of amino acids, nucleotides, and lipids via aerobic glycolysis. A key control for metabolizing glucose by either processes is to regulate the flux of glycolysis. Pyruvate kinase (PK) catalyzes the final reaction in glycolysis by transferring the high-energy phosphate from phosphoenolpyruvate (PEP) to ADP to produce ATP and pyruvate (Fig. 1) [3]; pyruvate is subsequently reduced to lactate by lactate dehydrogenase (LDH) in the cytosol or directed for high output of ATP production via the respiratory chain (Fig. 1). Therefore, a reduction in PK activity causes a build-up of glycolytic intermediates which are redirected towards biosynthesis (Fig. 1). This concept is supported by an observation in yeast that low levels of PK activity caused PEP accumulation and PEP blocked glycolysis via inhibiting the glycolytic enzyme triosephosphate isomerase [4].

PKs are encoded by the *PKLR* (1q22) and *PKM2* (15q23) genes, with *PKLR* producing the L (PKL) and R (PKR) isoforms and *PKM2* generating the M1 (PKM1) and M2 (PKM2) isoforms [5,6]. The expression of PK isoforms is under tissue-specific and developmental regulations. PKM2 is the predominant PK in fetal tissues, which is replaced by PKR in red blood cells, PKL in the liver, and PKM1 in skeletal muscle, heart, and brain in adults [5,7–10]. PKM2 remains the dominant M isoform in most adult tissues [8–12], and is the major PK in proliferating and cancer cells [13]. PKM1 and PKM2 are derived from the *PKM2* gene by mutually exclusive splicing of exon 9 and 10, respectively [6]. By retaining exon 10, PKM2 possesses unique properties important in the reprogramming of cell metabolism. Active PKs consist of four subunits [14], and PKL, PKR, and PKM1 form stable tetramers [14,15]. The exon 9 region plays a major role in forming PKM1 tetramers [16,17]. The exon 10-encoded residues allow PKM2 to exist as inactive monomers, less active dimers, and active tetramers [13,15,18,19], with PKM2 tetramers promoting high ATP generation and PKM2 dimers initiating high biosynthesis rates (Fig. 1) [15,20]. Adoption of the dimeric or tetrameric state by PKM2 is subjected to multiple extensive layers of regulation to meet the physiological needs of proliferating and cancerous cells, and is controlled by oncogenes and tumor suppressors [21–25]. Dimeric PKM2 not only directly promotes aerobic glycolysis by redirecting glucose-derived carbons towards biosynthesis, but also indirectly supports the Warburg effect by regulating gene expression in the nucleus, a process that involves the protein kinase activity of PKM2 [26]. We will discuss these regulations and their roles in cancer metabolism.

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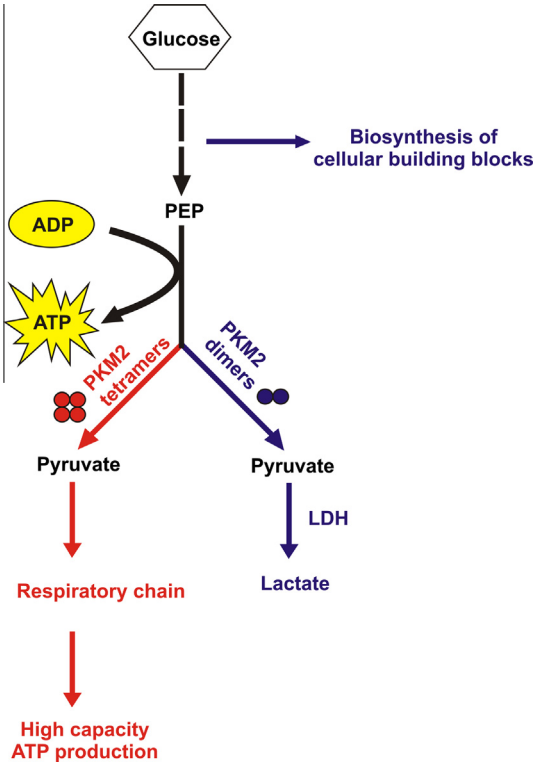

Fig. 1. A schematic illustration of the role PKM2 plays in reprogramming cancer metabolism. Pyruvate kinase catalyzes the last step of glycolysis by converting (phosphoenolpyruvate) PEP and ADP to pyruvate and ATP. PKM2 dimers and tetramers possess low and high levels of pyruvate kinase activity, respectively. By reducing the flux of glycolysis, the PKM2 dimer redirects glucose-derived carbons towards biosynthesis and the conversion of pyryuvate to lactate through lactate dehydrogenase (LDH); the PKM2 tetramer promotes the flux of glycolysis and high capacity of ATP production via oxidative phosphorylation through the respiratory chain.

2. Clinical evidence of PKM2 contribution to tumorigenesis

It was observed more than 40 years ago that liver cancer switched from PKL to PKM2 [8], a characteristic that is now predominantly seen in all cancers (Table 1). Elevated PKM2 expression has been demonstrated in the carcinoma of colon [27], renal cell carcinoma (RCC) [28] and lung [29]. PKM2 can be used as a marker for RCC and testicular cancer [30–33] and increases in serum PKM2 occur in cancer patients, including those of colon [33], breast [34], urological [35], lung, cervix, and gastrointestinal [13]. PKM2 was also detected in the feces of patients with gastric and colorectal cancer [36]. A unique pattern of four genes, including PKM2, was reported to predict outcomes for mesothelioma patients undergoing surgery [37]. Elevation of PKM2 associates with poor prognosis for patients with signet ring cell gastric cancer [38], esophageal squamous cell cancer [39], papillary thyroid cancer [40], small cell lung cancer [41], gallbladder cancer [42], and head and neck cancer [12]. In addition to PKM2, high levels of PKM1 was reported in the stroma of breast cancer [43].

Table 1
Elevation of PKM2 in human cancers.

Tumor with high levels of PKM2	Ref.
Breast cancer	[34]
Bladder carcinoma	[11,42]
Colon cancer	[27,11]
Esophageal squamous cell cancer	[39]
Glioma	[45,12]
Head and neck cancer	[12]
Liver cancer	[8,11]
Lung carcinoma	[29,11,41]
Meningiomas	[45,12]
Renal cell carcinoma	[28,30,11]
Renal oncocytoma	[11]
Thyroid cancer	[11,40]
Thyroid oncocytoma	[11]

During embryogenesis, the dominant fetal PK isoform PKM2 is gradually replaced by the tissue specific isoforms; PKL in the liver, and PKM1 in the brain, muscle, and heart [8,15,44]. PKM1 has the highest affinity for the substrate PEP and is constitutively active [15], thereby ensuring the energy supply for these tissues. For most other tissues, the dominance of PKM2 over PKM1 was recently reconfirmed [11]. While this reconfirmation does not support a shift from PKM2 over to PKM1 in tumors derived from these tissues [11], a shift could occur in tumors derived from tissues with dominant expression of other PK isoforms. In the PKM1 expressing brain, expression of PKM2 was reported in gliomas and meningiomas in 1977 [45], an observation that was recently confirmed [12]. During liver tumorigenesis, a decrease in PKL with a concomitant increase of PKM2 was reported in 1972 [8]. Despite in the latter situation where a shift was not seen between two M isoforms, it seems clear that cancer ensures the expression of PKM2. Following this logic, it is tempting to suggest that a shift over to PKM2 from PKM1 may occur in the rare cardiac and skeletal muscle tumors.

3. PKM2 promotes tumorigenesis by regulating the Warburg effect

Accumulating evidence reveals a critical role of PKM2 in tumorigenesis via promoting the Warburg effect. Knockdown of PKM2 in a panel of cancer cell lines reduced glucose uptake, increased oxygen consumption, and decreased lactate production; these changes were reversed when PKM2 was reintroduced, but not PKM1 [27]; re-expression of PKM2 promoted cell proliferation and xenograft tumor formation compared to cells in which PKM1 was re-expressed [27]. Regulation of the glycolytic activity of PKM2 is most likely critical for aerobic glycolysis, with the most potent small molecule PKM2 inhibitors reducing the Warburg effect in a manner that mimics the effects of PKM2 knockdown [46]. On the other hand, constitutively active PKM2 by a small molecule activator DASA-58 reduced lactate production and tumorigenesis [47], effects that were similar with the replacement of PKM2 with PKM1 [27]. Activating PKM2 by small molecule activators was reported to render cell proliferation dependent on serine, consistent with the knowledge that active PKM2 reduces anabolic metabolism [48,49]. Collectively, evidence supports a threshold level of PKM2 glycolytic activity being critical to aerobic glycolysis.

The inclusion of exon 10 enables PKM2 to be allosterically regulated by binding to an upstream intermediate, fructose-1,6-bisphosphate (FBP) [15,50], resulting in stabilization of the PKM2 tetramer in an active state (R-state) [51,52,50,19]. This allosteric regulation is also utilized by other mechanisms to regulate the equilibrium between the relatively inactive PKM2 dimers and the active tetramers (Tables 2 and 3) and small molecule PKM2 activators were produced to stabilize the PKM2 tetramer by binding to sites distinct from that of FBP [47].

3.1. PKM2 regulation

3.1.1. Post-translational modification of PKM2 regulates its activity and the Warburg effect

PKM2 was tyrosine-phosphorylated by v-Src, resulting in a decrease of PKM2's affinity towards its substrate PEP [53]. This early study is consistent with the recent demonstration of PKM2