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# Pre-Setting the Metabolic Pathways During Stem Cell Programming

Sai Sudha Purushothaman

## ABSTRACT

The role of metabolism has traditionally been viewed solely as a pathway for energy production without any influence on cell fate determination. However, in 1926, Otto Warburg reported that cancer cells exhibit high glucose consumption, popularly known as the Warburg effect. Cells orchestrate their metabolic program to meet their functional needs. All proliferating cells, from tumor cells to pluripotent cells such as naive, primed, and induced pluripotent cells, activated T and B cells shunt glucose to glycolysis and inhibit oxidative phosphorylation. This tailoring utilizes glucose in a biosynthetic pathway to generate metabolites that help in biomass production during cell division. Differentiated cells, designed to perform functional work, need more energy and hence follow oxidative phosphorylation to produce 36 molecules of adenosine triphosphate (ATP). Several studies have confirmed the direct link between glycolysis and cell proliferation. Pyruvate kinase M2 (PKM2) isoform enables dividing cells to adapt to a glycolytic lifestyle and promote anabolism. Stem cell transcription factors such as OCT4 and c-Myc promote glycolysis, while the AMP/ATP ratio functions as a sensor to determine the choice between glycolysis and oxidative phosphorylation. Tumor cells adhere to their own metabolic cues to promote tumorigenesis. Additionally, metabolic reprogramming produces acetyl CoA, lactyl residues, and S-adenosyl methionine, which contribute to epigenetic modifications and facilitate nuclear reprogramming.

**Keywords:** Metabolic reprogramming, Warburg effect, Pluripotent stem cells, Glycolysis, Pyruvate kinase M2

## Introduction

A long-standing hypothesis about the metabolic behavior of cancer cells was put forward by Otto Warburg in 1926. Warburg, a Nobel laureate for his research on respiratory enzymes, made a remarkable observation that cancer cells consume more glucose than normal cells from which they originated. He reasoned that increased glucose consumption was probably due to compromised mitochondria during tumorigenesis. As a result, cancer cells rely on the energy-inefficient

metabolic pathway, glycolysis, which is usually observed in the absence of oxygen, i.e., fermentation.<sup>1,2</sup> However, considering these proliferating cells adopt a glycolytic pathway even in the presence of abundant oxygen, the phenomenon was coined as aerobic glycolysis or the Warburg effect.

During that period, with limited tools, Warburg's theory on high glucose consumption by cancer cells due to mitochondrial damage may have failed.<sup>3–5,15</sup> However, his observation that cancer cells consume high glucose does stand the test of time as it is the basis for the FDG-PET scan, a cancer diagnostic tool (Figure 1). 2-Fluorodeoxyglucose (FDG) is a nonmetabolic, fluorescent analog of glucose with a –H group in the 2nd position rather than –OH as in glucose. Because FDG cannot be metabolized, it accumulates in the cells. Cancer cells rapidly consume FDG, resulting in greater accumulation in tumor cells than normal cells, and the observed fluorescence is directly proportional to the tumor size.<sup>6</sup> Renewed interest in cancer metabolism in the early 2000s shed light on the changing metabolic dynamics depending on the cell fate. Adaptation to aerobic glycolysis is not unique to cancer cells but is observed in all proliferating cells.<sup>7</sup> In this narrative review, we delve into the various metabolic fates of glucose in cells of different fates.

## Glucose: The Staple Diet Across All Scales

All cells need energy in the form of ATP for performing various biological activities such as ion transport across membranes, deoxyribonucleic acid (DNA) and ribonucleic acid (RNA) biosynthesis, intracellular signaling, and synaptic signal transmission. Glucose is the preferred molecule for energy production in different cells, from bacteria to fungi to plants and animals. In unicellular organisms, this energy demand is directly proportional to the availability of nutrients.<sup>8</sup> In contrast, in multicellular organisms with access to abundant nutrients, the metabolic rate is defined by both nutrients and growth factors (Figure 2). In mammals, glucose uptake is triggered by growth factors

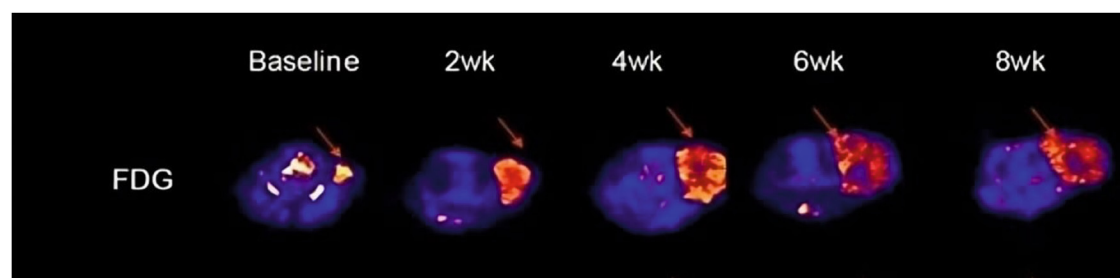


Fig 1 | Time course imaging of MNU-induced tumors at baseline, 2 weeks, 4 weeks, 6 weeks, and 8 weeks FDG-PET Scan. Shoghi et al., 2013. <https://doi.org/10.1371/journal.pone.0074188>

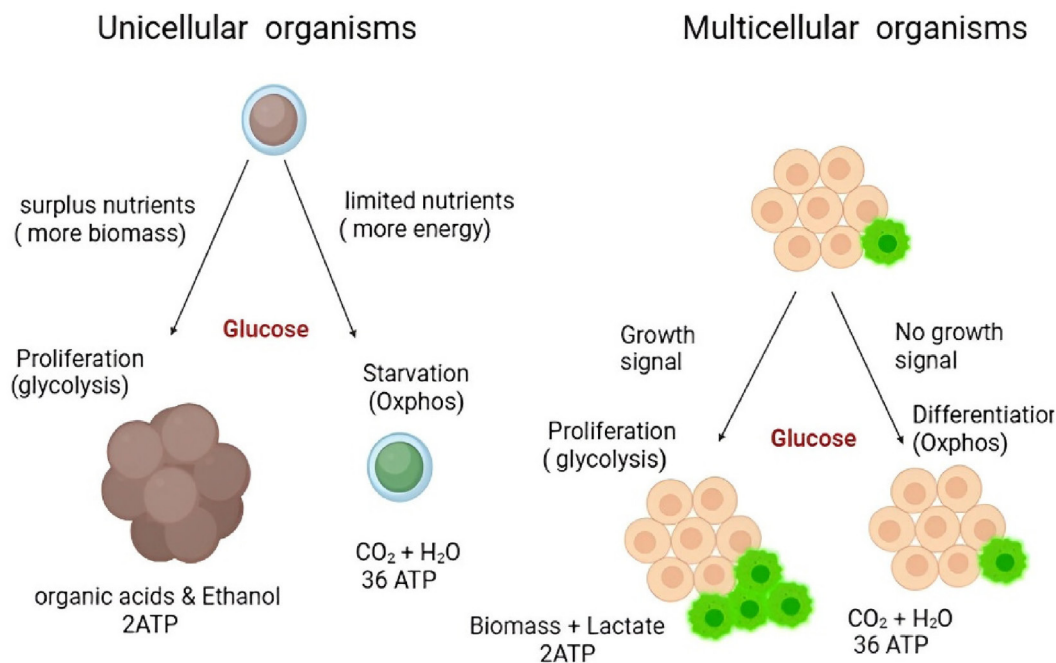


Fig 2 | Nutrient-sensing in unicellular organisms and nutrient- and growth-factor-sensing in multicellular organisms. Inspired by: Vander Heiden, 2009. <https://doi.org/10.1126/science.1160809>

such as insulin and epidermal growth factor (EGF). These factors activate receptor tyrosine kinases such as insulin receptor/insulin growth factor receptor (IR/IGFR) and epidermal growth factor receptor (EGFR), respectively, initiating a cascade of signalling events such as phosphoinositide -3- kinase (PI3-K) and protein kinase B (AKT) that facilitate glucose uptake.<sup>8,9</sup>

#### Energy Production Versus Biomass Accumulation

Cell proliferation needs the accumulation of macromolecules, such as proteins, nucleic acids, and lipids, in addition to DNA replication. The generation of biomass must sync with DNA duplication for cell division to occur effectively.<sup>8</sup> The intermediary metabolites in biomass production are provided by glycolysis, the hexose monophosphate (HMP) shunt, and the pentose phosphate pathway (PPP). In contrast, nondividing cells do not accumulate biomass but require energy to perform work. For example, skeletal myocytes require a large amount of energy to do work, and this demand is met by oxidative phosphorylation, which produces 36 molecules of ATP, the cell's energy currency. This raises an intriguing question: how do cells swing between producing more energy versus more biomass? This is particularly interesting since glucose, as one universal molecule, fulfills both requirements.

#### The Energy Paradox

Glucose metabolism for ATP production features several mechanisms. For example, *Saccharomyces cerevisiae* employs different metabolic pathways depending on its environment and nutrient supply: (a) under limited oxygen supply and excess glucose, yeast ferments to produce ethanol and ATP;<sup>10</sup> (b) as a regulatory process, under excess oxygen and glucose, it still

produces ethanol and ATP by a metabolic process similar to aerobic glycolysis called the Crabtree effect;<sup>11</sup> and (c) under excess oxygen and limited glucose, yeast derives its energy from oxidative phosphorylation.<sup>12</sup> This demonstrates that cells have devised multiple metabolic pathways for survival. The Warburg effect was observed in proliferating cells parallel to the Crabtree effect. This raises the question: Is the Warburg effect a regulatory process to adapt to the demands of higher biomass?

#### Warburg Effect: The Conceptions and Misconceptions

Proliferating cells adopt aerobic glycolysis, characterized by the preferential metabolism of glucose to lactate despite available oxygen. It is surprising that proliferating cells prefer glycolysis over oxidative phosphorylation in spite of less ATP production per molecule of glucose.<sup>7,13</sup> There is a clear distinction between the metabolic choices of proliferating and differentiated cells, with a bias toward biosynthetic or anabolic pathways in proliferating cells and a bioenergetic or catabolic pathway in differentiated cells.<sup>14</sup> In his observation, Warburg stated that cancer cells rapidly consume glucose, with most of the glucose directed into the glycolytic pathway (90%) in the cytoplasm, releasing lactic acid. In comparison, only 10% enters the mitochondria to be processed via the tricarboxylic acid (TCA) cycle and electron transport chain to undergo oxidative phosphorylation to release CO<sub>2</sub> and H<sub>2</sub>O (Figure 3). Warburg believed that cancer cells carried mutations in the mitochondrial respiration pathway, thus forcing them to rely on glycolysis.<sup>15</sup> However, after decades, in the early 2000s, it was evident that respiration was not impaired in most cancer cells; however, aerobic glycolysis was still preferred because of the advantages that it

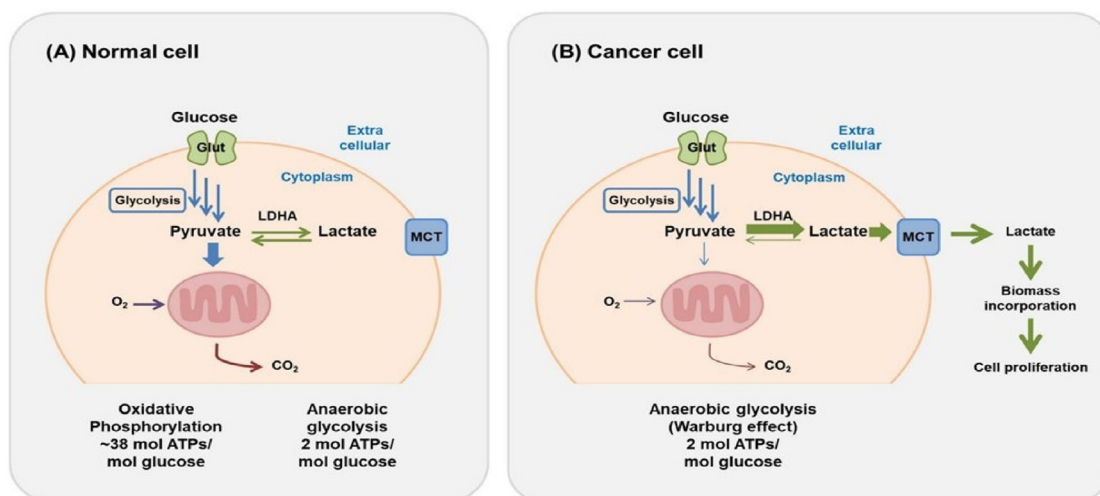


Fig 3 | Difference in glycolytic pathway between normal and cancer cells. Kim and Beak, 2021. [https://upload.wikimedia.org/wikimedia/commons/b/ba/Differences\\_in\\_glycolysis\\_pathways\\_between\\_normal\\_cells\\_and\\_cancer.webp](https://upload.wikimedia.org/wikimedia/commons/b/ba/Differences_in_glycolysis_pathways_between_normal_cells_and_cancer.webp)

provides for these proliferating cells. The question that interests us is the disparity in the energy demand between proliferating and differentiated cells.

Proliferating cells benefit by diverting glucose to alternate metabolic pathways beyond just ATP production. They adopt a mechanism that shuttles the flow of glucose into the pentose phosphate pathway to generate biosynthetic precursors to promote biomass accumulation. The Warburg effect takes advantage of faster ATP production by substrate-level phosphorylation, and also the production of important precursor molecules via the pentose phosphate pathway. Conversion of glucose to lactate in glycolysis results in carbon loss but increases the nicotinamide adenine dinucleotide ( $\text{NAD}^+$ ) pool during the reduction of pyruvate to lactate by lactate dehydrogenase.<sup>16</sup> When  $\text{NAD}^+$  becomes limited in the cytoplasm, glycolysis will stall as it is an electron acceptor in the conversion of glyceraldehyde-3-phosphate to 1,3-bisphosphoglycerate. Also,  $\text{NAD}^+$  is required for serine biosynthesis in highly proliferative cells.

### The Metabolic Landscape

Cells process glucose via catabolic and anabolic pathways depending on specific requirements (Figure 4). Catabolic pathways include aerobic or anaerobic respiration, which breaks down glucose by a series of enzymatic steps to produce ATP and other metabolic intermediates. In aerobic respiration, also known as oxidative phosphorylation, glucose in the presence of oxygen undergoes complete combustion to yield carbon dioxide and water, releasing 36 molecules of ATP. Initially, glucose is metabolized through glycolysis to form pyruvate, which enters the TCA cycle as acetyl CoA. This process generates electron-rich NADH, which produces ATP in the electron transport chain by oxidative phosphorylation.<sup>17</sup> Conversely, without oxygen, glucose can end in organic acids or alcohol, such as three-carbon pyruvate or lactate and 2-carbon acetate or ethanol, releasing two molecules of ATP during

anaerobic respiration or fermentation. Glucose can also enter the anabolic process via the pentose phosphate pathway to provide cells with reducing equivalents such as NADPH and ribose sugars essential for biomass production. NADPH molecules maintain the redox state of the cell and are also essential for fatty acid biosynthesis.<sup>18</sup>

The dynamics of cellular metabolic networks are synchronized to meet the specific demands of cell types. Cells of different lineages adapt to different metabolic patterns based on their function and niche.<sup>19</sup> Proliferating, differentiated, and quiescent cells have unique metabolic fingerprints. Naïve human pluripotent stem cells (hPSCs) follow oxidative phosphorylation to produce high ATP molecules, while primed hPSCs adopt aerobic glycolysis. In fact, factors that activate glycolysis promote induced PSC (iPSC) reprogramming.<sup>20</sup> Pyruvate from glucose is directed to mitochondria, where it undergoes decarboxylation to form acetyl CoA required for histone acetylation, essential for maintaining pluripotency. Surprisingly, the traditional catabolic pathway – TCA cycle supports the biosynthetic pathway in PSCs.<sup>21</sup> Excess acetyl CoA exits mitochondria into the cytoplasm as citrate. ATP-citrate lyase breaks citrate into acetyl CoA, precursors of fatty acid biosynthesis, and also molecules for epigenetic modification.<sup>22</sup> Within the stem cell population, different cell lineages carry specific metabolic choices. Naïve mouse embryonic stem cells (mESCs) rely on glycolysis and oxidative phosphorylation.<sup>23</sup> In contrast, primed mouse epiblast stem cells (mEpiSCs) are highly glycolytic with very little pipeline towards oxphos. Human ESCs (hESCs) from inner cell mass behave more like primed rather than naïve pluripotent cells and use glycolysis for metabolic demands.<sup>23</sup> Quiescent cells have a metabolic profile that is ever-changing in accordance with cell lineage, cell cycle, and their microenvironment. For instance, prolonged quiescent cells (oocytes), transiently quiescent cells (T cells), and permanently quiescent (senescent) cells have distinct



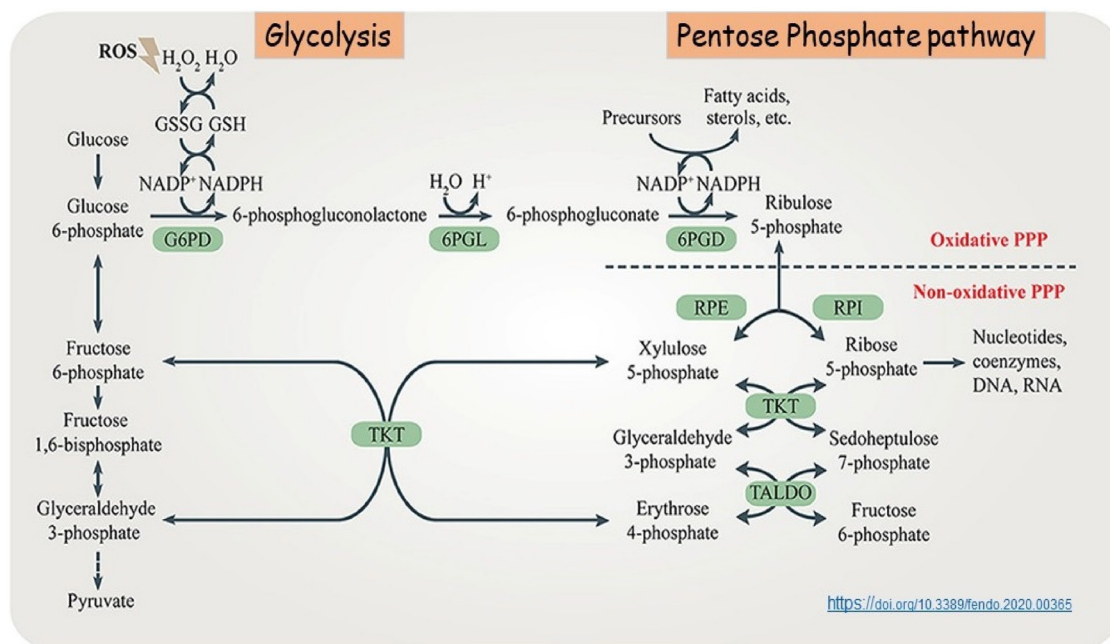


Fig 4 | Metabolites produced via glycolysis and pentose phosphate

metabolic strategies.<sup>24</sup> During embryogenesis from zygote to development into a multicellular organism, the cells traverse a changing metabolic landscape.<sup>25</sup> This proves that cellular metabolism is plastic and can be tuned to determine cell fate.

#### Metabolites That Regulate Proliferation

Metabolites from glucose breakdown have several critical importance; glucose-6-phosphate can be processed by glycolysis or shunted into the pentose phosphate pathway to produce ribose sugars and NADPH.<sup>19,21</sup> 3-phosphoglycerate is a precursor for amino acids such as glycine and serine.<sup>27</sup> The influence of glycolytic intermediates on nucleotide biosynthesis is particularly significant. Two of the five carbon atoms in purine are from glycine derived from 3-phosphoglycerate. The other two carbon atoms of the purine molecule are indirectly dependent on serine or glycine, which is a precursor for N10-formyl-tetrahydrofolate that contributes to the purine carbon skeleton.<sup>26,27</sup> This illustrates that ATP, guanosine triphosphate (GTP), cytidine triphosphate (CTP), and Thymidine triphosphate (TTP) depend on intermediates of the glycolytic pathway. Therefore, glycolysis and pentose phosphate pathways provide precursors for both DNA and RNA biosynthesis.

Aldolase breaks six-carbon glucose into dihydroxyacetone phosphate and glyceraldehyde-3-phosphate, which participate in the biosynthesis of phospholipids, glycolipids, and triacylglycerol which are major components of cell membranes. 3-phosphoglycerate, a downstream product of glyceraldehyde-3-phosphate, serves as a precursor for several amino acids, such

as cysteine, glycine, serine, and alanine.<sup>19,26</sup> Transketolase (TK) connects the glycolysis and pentose phosphate pathway to produce ribose sugars and NADPH, a redox molecule critical for fatty acid biosynthesis. Glycolysis-derived pyruvate is processed in the TCA cycle to generate acetyl CoA, the precursor of fatty acid biosynthesis. So, the foundation for glycolysis in proliferating cells is to enhance intermediate biomolecules required for faster cell biomass turnover.

Tetrahydrofolate can integrate carbon atoms from different metabolites at different positions of the moiety. Serine derived from 3-phosphoglycerate donates the side chain carbon to tetrahydrofolate (THF) to form glycine and methyl-THF, which forms bioactive molecules such as S-adenosyl methionine, which is involved in DNA methylation, histone methylation by one-carbon metabolism.<sup>28</sup> Thus, cells, by diverting glucose into different pathways, can manufacture important macromolecules to facilitate cellular proliferation.

#### Cellular Metabolism and Pluripotency

The transition of the metabolic profile from oxidative phosphorylation to glycolysis is required to induce pluripotency in somatic cells. The cells must make these remarkably quick and frequent choices between aerobic glycolysis and oxidative phosphorylation. ATP/AMP ratios are the input for the commitment to proliferation versus quiescence or differentiation.<sup>28</sup> Studies have shown that when glucose levels are low, the ATP turnover becomes low, resulting in elevated AMP levels, activating AMP-activated protein kinase (AMPK), which phosphorylates tumor suppressor p53, leading to G1-S cell cycle arrest.<sup>29</sup> Many of the glycolytic genes

are also regulated by stem cell transcription factors. In murine pluripotent stem cells (mPSCs), Octamer Transcription Factor 4 (OCT4) regulates the expression of hexokinase 2 and pyruvate kinase M2 (PKM2) to prevent differentiation. c-myc can upregulate the expression of PKM2 and lactate dehydrogenase (LDH) which in turn alleviates glycolysis.<sup>30</sup> Krüppel-like factor-4 (KLF4) expression is increased in reprogramming, but its function in metabolic regulation is yet to be confirmed.<sup>31</sup> In a feedback regulatory process, PKM2 positively upregulates the expression of OCT4, confirming the direct association between aerobic glycolysis and pluripotency in mPSCs. Silencing glucose transporter, GLUT3 suppressed the expression of OCT4, clearly representing a feedback control between glucose metabolism and expression of pluripotent transcription factors.<sup>32</sup> Morphologically somatic cells, mouse embryonic fibroblasts (MEFs) carrying elongated, matured mitochondria with dense cristae, transform into immature mitochondria on induction of pluripotency.<sup>31</sup>

Experimental evidence proves the importance of glycolysis, such as high glucose supplement stimulated glycolysis, 2-deoxy-D-glucose (2DG), an inhibitor of glycolysis-restricted reprogramming.<sup>32</sup> Pyruvate dehydrogenase (PDH) commits glucose into oxidative phosphorylation, and its phosphorylation by pyruvate dehydrogenase kinase (PDK) inhibits its activity and commits the cells to glycolysis. Studies show that dichloroacetate (DCA), an inhibitor of PDK, reduces reprogramming efficiency, as an active PDH shuttles pyruvate to TCA and concomitantly to oxidative phosphorylation, thereby suppressing glycolysis.<sup>33</sup> The metabolic intermediate of glycolysis can choose different paths to satisfy the high anabolic requirements of proliferating cells.<sup>34</sup>

Stimulation of quiescent mouse T cells by antigens leads to elevated expression of glucose transporter-3 (GLUT-3), causing substantial glucose uptake for aerobic glycolysis and proliferation of activated mouse T cells.<sup>35</sup> This is in contrast to resting lymphocytes, which exhibit little glucose. The glycolysis releases 3-phosphoglycerate, which functions as a precursor for amino acids. Also, dihydroxyacetone phosphate and acetyl CoA, intermediates of glucose metabolism, are utilized for lipid synthesis, and glucose-6-phosphate enters the pentose phosphate pathway to produce nucleotides. All these metabolites help in building the infrastructure for cell division.

### Metabolic Reprogramming Drives Induced Pluripotency?

Transformation of differentiated cells to a pluripotent state requires transcriptional, epigenetic, and metabolic reprogramming. The focus has been on transcriptional reprogramming with well-defined roles for Yamanaka factors to transit differentiated cells to a pluripotent state. Metabolic reprogramming to aerobic glycolysis precedes nuclear reprogramming, which involves the induction of pluripotent markers. Fructose-6-phosphate (F6P), an activator of glycolysis,

seems to influence the conversion of human fibroblasts to iPSC, as observed by the increase in the number of iPSC colonies. 2-deoxy-D-glucose (2-DG), an analog of glucose inhibits glycolysis, thereby decreases reprogramming efficiency. Overexpression of HIF1 $\alpha$ , a transcription factor that ups the expression of glycolytic genes, improves reprogramming efficiency.<sup>36</sup> Also, cells such as keratinocytes, more adapted to glycolytic lifestyle, can be more efficiently reprogrammed. These studies illustrate the central role of metabolism in reprogramming. The field of induced pluripotency utilizes small molecules to activate transcription factors to promote reprogramming. In a similar fashion, PS48, a small-molecule PDK1 activator which increases glycolytic gene expression along with other small molecules, functionally replaces SOX2, KLF4, and c-Myc in reprogramming keratinocytes.<sup>37</sup> These studies prove that presetting metabolic programs from oxidative phosphorylation to glycolysis can be exploited for driving reprogramming. Metabolic and epigenetic reprogramming may be the next-generation targets in cellular reprogramming.

### Pyruvate Kinase, the Central Player in Pluripotency

Pyruvate kinase M2 (PKM2) stands between adaptation to glycolysis and oxidative phosphorylation, equating with cell fate determination between pluripotency and differentiation. Pyruvate is a central molecule that regulates the fate of glucose, either terminating in aerobic glycolysis on reduction to lactate or enter mitochondria to get oxidized to carbon dioxide and water. Pyruvate kinase (PK) is an allosteric enzyme that limits the rate of turnover of glycolysis. The enzyme catalyzes the conversion of phosphoenolpyruvate to pyruvate coupled with the production of ATP from ADP and Pi. In mammals, PK has different isoforms depending on tissue specificity and allosteric regulation. The four isoforms are expressed by two genes: *PKLR*, which is expressed in the liver and red blood cells; and *PKM*, which is expressed in various tissues by alternate splicing. Pyruvate kinase M isoform is again of two types: constitutively active PKM1 expressed in differentiated cells; and PKM2 allosterically activated by Fructose-1,6-bisphosphate (FBP) expressed in proliferating cells. PKM1 and PKM2 are isoforms formed by alternative splicing: PKM1 includes exon nine, while PKM2 is formed by the inclusion of exon ten. PKM1 exists as a tetramer, while PKM2 can take different oligomeric forms, such as monomer, dimer, or tetramer allosteric, by allosteric binding of FBP, phosphotyrosine molecules, and nuclear localization signals (NLS). On binding to phosphotyrosine molecules, PKM2 releases FBP, making the enzyme functionally weak and thereby facilitating the flux of glucose into the anabolic pathways, from the traditional catabolic pathways. Due to its slow enzymatic activity, the glucose flux is shunted to the pentose phosphate pathway to form other macromolecules required for biomass production. Christofk et al<sup>38</sup> showed that the expression of M2 isoform of pyruvate kinase in cancer cells presets the switching to aerobic glycolytic phenotype or Warburg effect.<sup>39,40</sup>

PKM2 exists in two different oligomerization states determined by FBP and phosphotyrosine. The higher oligomeric PKM2 (tetramer) is enzymatically more active than the monomeric or dimeric form. FBP promotes the tetramerization of PKM2, which has a higher affinity for PEP to produce pyruvate. However, in the presence of phosphorylated proteins, the enzyme FBP dissociates, leading to a lower oligomeric state of PKM2 with reduced affinity for PEP.<sup>41</sup> As the affinity of PKM2 for PEP reduces, glucose-6-phosphate accumulates, which then enters alternate pathways such as pentose phosphate, hexosamine, and glycerol biosynthesis pathways. These pathways generate important precursor molecules required for biomass production. C-Myc, an oncogene, has been found to promote preferential expression of PKM2 over PKM1 by modulating exon splicing. Switching between biosynthetic and bioenergetics pathways is primarily regulated by the splice variant of PKM2 and its propensity to alter different oligomeric forms.

#### Metabolic Reprogramming and Macrophages

Metabolic reprogramming is not restricted only when cells change their fate from proliferation and differentiation, but also during the activation of immune cells, such as macrophages. Macrophages can adapt to two different types: the M1 subtype induced by lipopolysaccharide (LPS), IFN- $\gamma$  releases a pro-inflammatory response, and the M2 subtype induced by IL-4, IL-10, and IL-13 releases an anti-inflammatory response. Reflecting on their metabolic choices, macrophages execute a high degree of plasticity, M1 follows aerobic glycolysis, and M2 follows oxidative phosphorylation. This selective metabolic preference in different macrophage phenotypes can be therapeutic targets to toggle between pro-inflammatory and anti-inflammatory responses.

Another interesting and surprising milestone in epigenetics is lactylation, a novel post-translational modification (PTM) of histone proposed by Zhang et al.<sup>42</sup> Accordingly, lactate accumulated by aerobic glycolysis or from the extracellular environment promotes lactylation of lysine residues of histone and nonhistone proteins. While histone acetylation alters chromatin remodeling, resulting in transcriptional regulation of various genes, nonhistone lactylation regulates stability, intermolecular interactions, and localization of the modified protein. Thus, lactate functions as an important metabolite in macrophages that regulates its phenotype through lactylation modification.

#### Cellular Metabolism and Cancer

There is a direct regulation between carbon metabolism and tumor-specific genes. For instance, oncogenes such as c-Myc and Kirsten rat sarcoma viral oncogene homolog (KRAS) enhance the expression of several genes, such as glucose transporter-1 (GLUT-1), to promote glycolysis in tumour cells. In inverse regulation, tumour suppressors such as TP53 suppresses GLUT-1 expression.<sup>43</sup>

As we are already aware, glycolysis is the preferred metabolic pathway adopted by cancer cells. A simplistic drug would be an analog of glucose, 2-deoxy

glucose (2-DG), which cannot be metabolized in the cells, and thus, the cells will be deprived of glucose for proliferation. Though this drug seemed promising for its antiproliferative effects, it failed because it induced severe hypoglycemia in patients. Another therapeutic target is isocitrate dehydrogenases 1 (IDH 1) and 2 of the citric acid cycle. Mutations in these genes have been reported in glioblastomas. These mutants produce D-2-hydroxyglutarate(D-2HG), an analog of  $\alpha$ -ketoglutarate, which inhibits  $\alpha$ -ketoglutarate-dependent dioxygenases and enzymes involved in DNA methylation, resulting in histone hypermethylation, thereby promoting oncogenesis. This makes IDH1 and IDH2 potential metabolic targets for cancer treatment. AGI-5198 and AG-221 are small-molecule inhibitors targeting IDH1 and IDH2, respectively. Other anticancer drugs that target mutant IDH1 include AG-120, IDH305, and AG-881. Another area of drug target is glutaminolysis, as most cancer cells are addicted to glutamine. Also, succinate dehydrogenase (SDH) and fumarate hydratase (FH) have been shown to possess tumor-suppressing properties.<sup>44</sup>

In tumor microenvironment (TME), there is a conflict between tumor-infiltrating T-lymphocytes (TILs) and tumor cells for nutrients. Under this condition, the T-cell metabolism is compromised, resulting in the failure of effector function, which affects its ability to clear tumor cells. Also, interaction of programmed death 1 (PD-1) and cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) with respective checkpoint molecules on tumor cells impairs glycolysis by suppressing the PI3-K/AKT/mTOR signaling pathway, thereby enhancing the immunosuppressive activity of T cells. In this section, we explore the aberrant metabolic processes that contribute to the depletion of CD8+ T cells within TME. Nutrient depletion and accumulation of certain metabolic by-products alter the metabolic activities of CD8+ T cells, which impairs their ability to produce IFN- $\gamma$ .<sup>44</sup> On prolonged deprivation of nutrition, CD8+ T cells produce inhibitory cytokines, which leads to permanent dysfunction of these cells.<sup>45</sup>

#### Future Perspectives on Metabolic Reprogramming

Considering that tumor cells follow specific metabolic pathways, novel therapies targeting these pathways would provide a more effective tumor elimination. Aerobic glycolysis, glutamine addiction, and oncometabolites determine tumorigenesis and, hence, are major drug targets. It has also been shown that immune cells follow their own trademark metabolic pathways and hence may be activated by using molecules that trigger specific metabolic pathways that activate immune cells for tumor clearance. Cell metabolism is a key player in cell fate determination during pluripotency, differentiation, and induced pluripotency. By tuning metabolic pathways, we can probably enhance tissue regeneration and stem cell differentiation.

#### Conclusion

This review provides a snapshot of the direct link between metabolic choices and cell fate. It also explores the key regulators and sensors that switch the metabolic fate of glucose during induced pluripotency.



Proliferating cells benefit from a biosynthetic or anabolic pathway to produce metabolites that enhance biomass production. In contrast, differentiated cells prefer bioenergetics or catabolic pathways to harvest a maximum yield of 36 ATP molecules. In proliferating cells, PKM2, an isoform of pyruvate kinase, slows glycolysis to shunt glucose into the pentose phosphate to produce ribose sugars and redox molecules such as NADPH. The review reiterates that metabolic reprogramming precedes nuclear programming, suggesting a primordial role of metabolism in cell programming. We need to explore if metabolic reprogramming by small molecules could be an alternate means of induced pluripotency. Also, targeted metabolic pathways could be the next-generation anticancer drugs.

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