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Chicken or the egg: Warburg effect and mitochondrial dysfunction

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Abstract

Compared with normal cells, cancer cells show alterations in many cellular processes, including energy metabolism. Studies on cancer metabolism started with Otto Warburg's observation at the beginning of the last century. According to Warburg, cancer cells rely on glycolysis more than mitochondrial respiration for energy production. Considering that glycolysis yields much less energy compared with mitochondrial respiration, Warburg hypothesized that mitochondria must be dysfunctional and this is the initiating factor for cancer formation. However, this hypothesis did not convince every scientist in the field. Some believed the opposite: the reduction in mitochondrial activity is a result of increased glycolysis. This discrepancy of opinions is ongoing. In this review, we will discuss the alterations in glycolysis, pyruvate metabolism, and the Krebs cycle in cancer cells and focus on cause and consequence.

Introduction

Almost a century ago, Otto Warburg made a very significant observation that would start a long-lasting, heated discussion. He observed that cancer cells, unlike many other cells in the body, opt for glycolysis rather than mitochondrial respiration, even in the presence of oxygen (referred to here as the Warburg effect) [1]. Warburg proposed that the aerobic glycolysis phenotype that he observed stemmed from the fact that cancer cell mitochondria are irreversibly dysfunctional. He believed, in fact, that dysfunctional mitochondria are required and necessary to start all the biochemical events that eventually result in transformation to the cancerous state [2]. His findings went hand-in-hand with Pasteur's postulations. In 1861, Pasteur reported that yeast cells upregulate glycolysis under hypoxic conditions. Given that the inner regions of solid tumors are hypoxic because of anomalous vascularization, Pasteur's effect seemed to explain Warburg's observation. However, the biochemist Weinhouse was not convinced by Warburg's explanation of cancer initiation by damaged mitochondria [3,4]. As a pioneer of isotope tracer usage in biochemistry, he found that cancer cells are able to oxidize glucose and fatty acids to carbon dioxide at levels comparable to those of normal cells [5]. He argued that the reverse was true: cancer cells have reduced mitochondrial activity as a consequence of heightened glycolytic flux, which is known to inhibit mitochondria—the so-called Crabtree effect [6,7]. To this day, the field has not been able to reach a conclusive decision on this matter. To explore the relationship between these two views, we use the chicken-and-egg analogy: it is difficult to determine whether mitochondrial dysfunction emerges first, thereby forcing cells to rely on glycolysis, or whether the reverse occurs, whereby increased glycolytic flux takes place first, which in turn suppresses mitochondrial respiration. There are data supporting both of the models in different contexts. In this review, we will discuss the two different points of view in relation to glycolysis, pyruvate metabolism, and the Krebs cycle.

Changes in the glycolytic pathway during tumorigenesis

As Warburg noticed, cancer cells have elevated levels of glucose uptake compared with non-cancer cells. These findings have been confirmed by using recent technological developments that allow non-invasive monitoring of glucose uptake *in vivo*, called 2-fluoro-6-deoxyglucose positron emission tomography (FDG-PET). In this technique, a traceable glucose analog is used that is

recognized and taken up by glucose transporters (GLUTs) but cannot be used for downstream glycolytic reactions. Hence, the glucose analog accumulates in cells and can be visualized and quantified by PET. This technique also allows diagnosis and localization of tumors. With FDG-PET, glucose uptake has been shown to increase in certain tumors [8], and higher FDG uptake has been correlated with poor cancer prognosis [9–11].

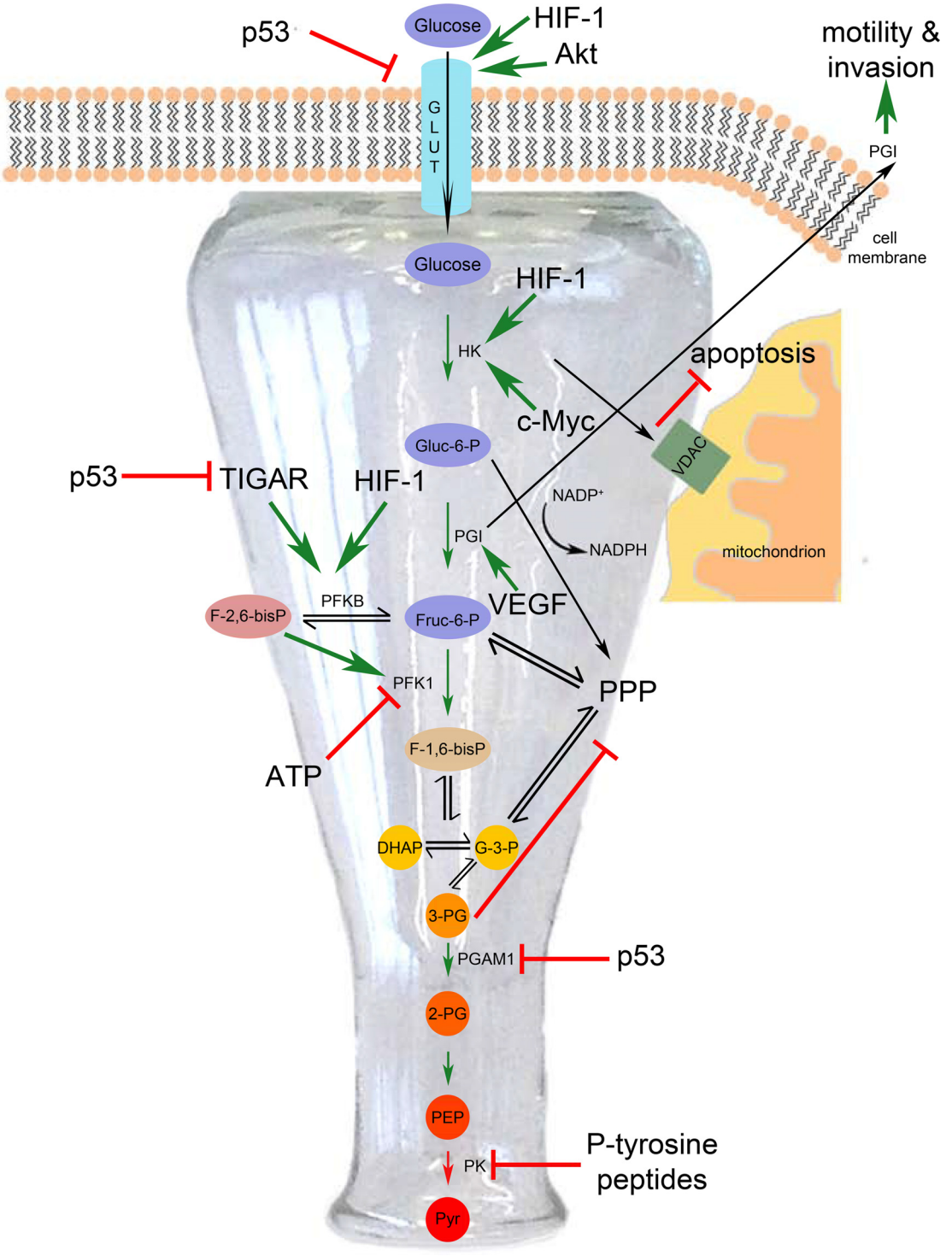
Multiple observations outlined below, however, suggest that glycolytic flux in cancer cells is upregulated as a result of multiple changes in signaling pathways and not necessarily as a result of impaired mitochondrial function. Glycolysis begins with cellular uptake of glucose via GLUTs on the cell surface (Figure 1). In 1974, Hatanaka proposed that cells upregulate GLUTs in order to meet increased glucose demand upon transformation [12]. Later studies showed that, indeed, levels of GLUTs, especially the high-affinity GLUTs 1 and 3, were upregulated in a plethora of tumor types (nicely reviewed in [13,14]). Moreover, *GLUT1* transcription is upregulated in response to hypoxia [15,16] and inhibition of mitochondrial respiration [17], both conditions in which cells need to divert the metabolic flux from mitochondrial respiration to glycolysis. Furthermore, in tumors with high insulin signaling, GLUT4 is enriched at the cell membrane as a consequence of elevated PI3K/Akt signaling (reviewed in [18]) and *GLUT1* transcription is upregulated via the serine/threonine kinase AKT [19]. The human genome actually has three families of GLUTs, namely SLC2A, SLC5A, and SLC50A, with a total of 27 members [20]. These members are differentially regulated in various tumor types. These findings could suggest that upregulation of glucose uptake and hence glycolytic flux is a primary alteration in cancer and not a consequence of impaired mitochondrial function. That said, activity of GLUTs is also strongly driven by activation of AMP-activated protein kinase (AMPK) [21], and AMPK can be activated by an adenosine triphosphate (ATP) decrease caused by mitochondrial dysfunction. Therefore, increased glucose uptake could also result from mitochondrial dysfunction. This nicely illustrates the fact that glycolytic flux and mitochondrial function are so intertwined that it is difficult to determine what is cause and what is consequence.

After uptake into the cell, the next step in glycolysis is phosphorylation of glucose to glucose-6-phosphate by hexokinase (HK) (Figure 1). There are four isoforms of HK and upon transformation, isoform II, the isoform with the highest enzymatic activity, becomes the prevalent isoform in the cell [22] and this is due in part to HIF1α (hypoxia-induced factor 1 α)-dependent transcriptional upregulation [23]. HKI and especially HKII are known to interact with voltage-dependent anion channels (VDACs) on the mitochondrial outer membrane of rapidly proliferating cells. This interaction is important for the inhibition of apoptosis by blocking cytochrome c release into the cytoplasm [24]. This helps cells evade apoptosis, one of the six hallmarks of cancer [25]. For these reasons, the VDAC-HKII interaction is a potential target for cancer therapy. There are data showing that the VDAC-HKII interaction favors glycolysis by inhibiting a negative feedback of HKII by its own product and by stabilizing HKII protein [26,27]. Thus, HKII offers a mechanistic explanation for the Warburg effect independently of mitochondrial dysfunction.

Glucose-6-phosphate is next converted to fructose-6-phosphate by phosphoglucose isomerase (PGI) (Figure 1). Expression of *PGI* is induced in response to HIF-1α and vascular endothelial growth factor (VEGF) signaling, both of which are often deregulated in tumors [28]. Interestingly, PGI acts as a cytokine outside of the cell. Secreted PGI is a tumor marker, as it can be detected in serum and urine of patients with cancer [29,30]. PGI is also called AMF (autocrine motility factor) because treatment of fibrosarcoma cells with purified PGI protein induces cell migration [31], implicating it in metastasis. Furthermore, mere PGI gain of function is sufficient to drive cell proliferation in 3T3 fibroblasts [32]. All in all, *PGI* can be considered an oncogene.

Next, fructose-6-phosphate is phosphorylated again to yield fructose-1,6-bisphosphate (Figure 1). This step is of particular importance for regulation of metabolism for several reasons. Firstly, it is the rate-limiting reaction. Secondly, the fact that ATP, its own substrate, allosterically inhibits phosphofructokinase 1 and 2 (PFK1 and 2) [33-35] offers an explanation for the Pasteur effect: inhibition of glycolysis by mitochondrial respiration. Thirdly, this is a decision point for glucose to enter further into glycolysis or to be diverted into the pentose phosphate pathway (PPP). The PPP has two branches: an irreversible, oxidative branch that starts with glucose-6-phosphate and generates reduced nicotinamide adenine dinucleotide phosphate (NADPH) and a reversible, non-oxidative branch that interconverts 5-carbon sugars and produces no NADPH (Figure 1). Hence, changes in PFK activity influence how much glucose enters the oxidative branch of PPP, yielding reducing equivalents for fatty acid biosynthesis and reactive oxygen species (ROS) defense (Figure 1). One critical factor determining PFK1 activity is the level of fructose-2-6-biphosphate, which is the product of the bifunctional enzyme phosphofructokinase 2/fructosebisphosphatase (PFKB). Fructose-2, 6-bisphosphate allosterically activates PFK1, thereby counteracting ATP inhibition [36,37]. *PFKB* has four

Figure 1. In cancer, glycolytic flux is increased through upstream parts of the glycolytic pathway up to pyruvate kinase and then decreased from pyruvate kinase downward, thereby generating a 'bottleneck'



DHAP, dihydroxyacetone phosphate; F-1,6-bisP, fructose-1,6-bisphosphate; Fruc-6-P, fructose-6-phosphate; Gluc-6-P, glucose-6-phosphate; GLUT, glucose transporter; HIF-1, hypoxia-induced factor 1; HK, hexokinase; PEP, phosphoenolpyruvate; PFK, phosphofructokinase; PG, phosphoglycerate; PGAM1, phosphoglycerate mutase 1; PGI, phosphoglucose isomerase; PK, pyruvate kinase; pyr, pyruvate; PPP, pentose phosphate pathway; TIGAR, TP53-induced glycolysis and apoptosis regulator; VDAC, voltage-dependent anion channel; VEGF, vascular endothelial growth factor.

isozymes that are expressed in a tissue-specific manner; however, *PFKBP3*, which has the highest kinase activity, is upregulated in high-grade astrocytomas and malignant breast and colon tumors [38,39], tipping the scale of the bidirectional reaction in favor of fructose-2,6-bisphosphate production. This sustains increased glycolytic activity rather than PPP. Upregulation of PFKB expression and activity occurs in various ways. A recent publication showed that methylation stabilizes PFKB3 in U937 human leukemia cells [40]. All PFKB isoforms are upregulated in response to hypoxia *in vivo*, *PFKBP3* being induced to the largest extent [41]. In addition to *PFKB3*, *PFKB4* has been also found to be important for cancer cell survival via small interfering RNA (siRNA) screens in two different models: glioma stem-like cells [42] and prostate cancer cell lines [43]. Another critical factor controlling PFK activity is the p53 tumor suppressor-mediated induction of *TIGAR* (TP53-induced glycolysis and apoptosis regulator). TIGAR negatively regulates PFK2 activity and lowers fructose-2,6-bisphosphate levels, and thereby reduces PFK1 activity and diverts glucose into the PPP rather than glycolysis [44]. In tumors with p53 loss of function, TIGAR thereby contributes to the increase in glycolytic rate. p53 has additional regulatory roles in metabolism, such as down-regulating *GLUT1* and *GLUT*4 expression at the transcriptional level [45].

Phosphoglycerate mutase 1 (PGAM1) is another glycolytic enzyme whose activity is increased in several tumor types, including hepatocellular carcinoma [46]. Its protein level is negatively regulated by p53 [47]. PGAM1 converts 3-phosphoglycerate to 2-phosphoglycerate (Figure 1). Its activity is important in metabolic regulation because its substrate, 3-phosphoglycerate, inhibits flux through the oxidative branch of PPP [48], thereby diverting glucose into glycolysis. Therefore, upregulation of PGAM1 is advantageous for cell proliferation, although it renders tumor cells sensitive to oxidative stress due to reduction in NADPH production from the oxidative branch of PPP.

Pyruvate: the intersection

So far, we have discussed the steps of glycolysis through which flux is increased in cancer cells. Interestingly, cancer cells do not increase flux in all steps of glycolysis. Although flux through the upstream steps is increased, they often exhibit a bottleneck in the following steps of glycolysis leading from phosphoenolpyruvate (PEP) to the Krebs cycle and this is for the reasons discussed below.

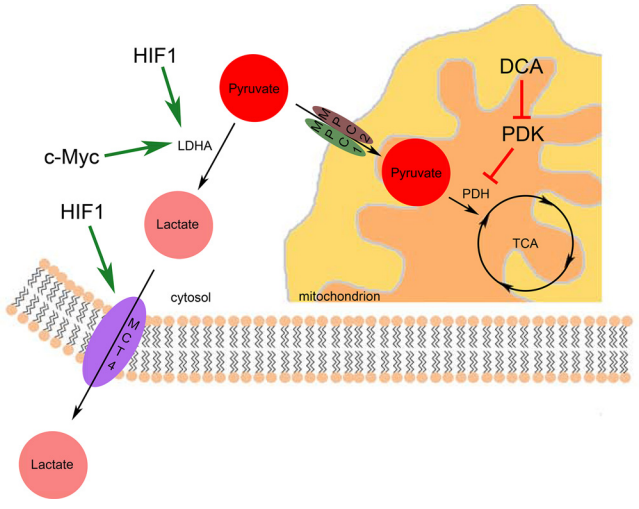
Pyruvate kinase (PK) is an important node of control in cancer cell metabolism. It catalyzes the conversion of PEP to pyruvate (Figure 1). PK has two isoforms produced by alternative splicing of *PKM*, namely M1, and M2, which are spatially and temporally differentially expressed. PKM1 is expressed mainly during adulthood, and PKM2 is expressed during embryogenesis. The alternatively spliced transcripts of *PKLR*, the L and R isoforms, on the other hand, are expressed specifically in liver [49]. In 2008, it was shown that expressing PKM2 confers a proliferative advantage to tumor cells [50]. In fact, depletion of PKM2 expression in cancer cell lines and reconstitution with PKM1 led to inhibition of growth and reversal of the Warburg phenotype [50]. This finding was unexpected because at first glance it does not fit into the model that cancer cells have increased glycolytic flux. PKM2 is the isoform with lower enzymatic activity and therefore it slows down this glycolytic reaction. However, generating a bottleneck at the end of the glycolytic pathway appears to be necessary for giving intermediates the time to flux through alternative pathways, such as PPP, that are of crucial importance for replenishing building blocks required for growth and proliferation [49]. PPP flux is especially needed in cancer cells for the production of pentose phosphates, which are used for nucleotide biosynthesis, and for the production of NADPH, which is a reducing equivalent needed both for fatty acid and sterol biosynthesis [51] and for mounting an anti-oxidant response to ROS by re-oxidizing glutathione [52]. PKM2 is also subject to negative regulation by phosphotyrosine peptides. Since tyrosine kinase signaling is often deregulated in a cancer setting, this regulation could be another way of pushing glycolytic intermediates into anabolic pathways such as the PPP [53]. Another advantage that selective PKM2 expression confers to cancer cells is the accumulation of its substrate, PEP. PEP acts as a phosphate donor to phosphorylate PGAM1 at the catalytic histidine, thereby increasing its activity [54]. Interestingly, the functional relevance of PKM2 in tumor development was analyzed in a recent study in which mice specifically lacking the PKM2 isoform were found to develop breast tumors and liver metastases induced by *BRCA1* loss of function, indicating that PKM2 per se is not required for tumor development [55]. Tumor cells had compensatory PKM1 expression, although PKM1 levels were heterogenous throughout the tumor. Whereas non-proliferating tumor cells exhibited higher PKM1 expression, proliferating cells did not [55]. The same study also analyzed human tumor samples and reported the presence of tumor samples with no detectable PK expression. Even though further studies are required, these findings hint that proliferating cells can lack PK activity altogether. It appears that PKM2 expression per se is not required. Rather, the outcome of reduced PK activity is what favors cell proliferation.

Pyruvate is the critical node where the flux of glucose-derived carbons is determined, either toward lactate which is usually secreted or into mitochondria (Figure 2). As Warburg observed, cancer cells metabolize pyruvate by aerobic glycolysis and produce lactate. Here, we will discuss the mechanisms of aerobic glycolysis induction which do not necessarily stem from dysfunctional mitochondria, which would have pleased Weinhouse.

The first possibility for pyruvate is to be reduced to lactate by lactate dehydrogenase (LDH). The prevalent isoform of LDH is LDHA. Like many other glycolytic genes, *LDHA* is induced by both HIF-1α [56] and c-myc [57]. Another means of inducing LDHA activity in the context of cancer is via phosphorylation at the tyrosine 10 residue [58]. Reflecting the importance of the Warburg effect for cancer cell survival, loss of function of LDHA by means of either siRNA depletion or pharmacological inhibition forces mitochondria to respire and slows down proliferation [59,60]. To maintain intracellular pH homeostasis, cells need to evacuate the resulting lactate to the extracellular space. Indeed, loss of function of the hypoxia-responsive lactate/H^+ symporter *MCT4* (monocarboxylate transporter 4) [61] impairs tumor growth [62], thereby rendering *MCT4* a good therapeutic target.

The second possibility of pyruvate utilization is to send it to mitochondria for further oxidation in the Krebs cycle.

Figure 2. In cancer cells, reduction of pyruvate to lactate and its secretion is favored rather than pyruvate entry into mitochondria and the Krebs cycle



DCA, dichloroacetate; HIF1, hypoxia-induced factor 1; LDHA, lactate dehydrogenase A; MCT4, monocarboxylate transporter 4; MPC, mitochondrial pyruvate carrier; PDH, pyruvate dehydrogenase; PDK, pyruvate dehydrogenase kinase; TCA, tricarboxylic acid.

The limiting step here is the entry of pyruvate into mitochondria by mitochondrial pyruvate carriers (MPCs) (Figure 2). The activity and chemical inhibition of a MPC were shown in 1974 [63], but the molecular identities of these carriers remained a secret until 2012, when they were identified as *MPC1* and *MPC2* [64,65]. The cancer relevance of these carriers was thoroughly investigated very recently [66–68]. In *MPC1* loss-of-function tumor models, forced expression of the protein leads to activation of mitochondrial pyruvate oxidation, which inhibits anchorage-independent growth of colon cancer cells [68]. These studies indicate that MPC loss of function could be one means of rewiring cancer metabolism toward aerobic glycolysis, the Warburg effect. In tumor types with reduced MPC activity, it could be used as a therapeutic target. The fact that forced activation of MPC can activate mitochondrial respiration suggests that mitochondria are not necessarily irreversibly damaged, as Warburg claimed. Upon entry into mitochondria, pyruvate is converted to acetyl coenzyme A (CoA) and joins the Krebs cycle, during which reduced nicotinamide adenine dinucleotide (NADH) and precursors for anabolic pathways are generated (Figure 3A).

At this stage, there is one more regulation point, pyruvate dehydrogenase (PDH). PDH is negatively regulated by NADH and acetyl CoA and through phosphorylation by pyruvate dehydrogenase kinase (PDK) [69,70]. PDK is a direct target of HIF1 [71,72]. HIF1 upregulates PDK transcription in response to hypoxia to reduce mitochondrial utilization of glucose. Moreover, oncogenic tyrosine kinases contribute to the reduction of PDH activity in two ways: they phosphorylate PDK to increase its activity [73] and they phosphorylate and inhibit a phosphatase that acts on PDH [74]. Dicholoroacetate (DCA) is a pharmacological inhibitor of PDK [75–78], which is used in treatment of lactic acidosis and cancer. The fact that DCA treatment is able to reverse Warburg phenotypes [76] is another indication that mitochondria do not have to be irreversibly damaged.

'Hacking' the Krebs cycle

Even though cancer cells tend to use glucose in aerobic glycolysis rather than oxidizing it in the Krebs cycle, the Krebs cycle is not dispensable altogether. The Krebs cycle constitutes a chain of reactions that are essential for the production of building blocks for growth and proliferation. As anabolic reactions use substrates from the Krebs cycle, thereby depleting the Krebs cycle, the cell needs to replenish these intermediates via a set of so-called anaplerotic reactions [79]. For example, Vacanti and colleagues [66] made the observation that, upon inhibition of pyruvate entry into mitochondria in C2C12 myotubes, cells were able to maintain Krebs cycle

metabolism without any drastic changes in the levels of intermediates, even though they exhibit a reduction in the mitochondrial oxidation of pyruvate. Cancer cells that divert glucose metabolism away from mitochondria face a situation similar to pyruvate entry inhibition and therefore employ strategies to keep the cycle going, discussed below. By this means, they use mitochondria for biosynthesis rather than degradation.

One of the anaplerosis strategies is glutaminolysis (Figure 3A). Cells use glutamine as a carbon source to feed the Krebs cycle [80]. Glutamine is first converted to glutamate by glutaminase (GLS). Next, glutamate is converted to α-ketoglutarate (α-KG) either by a transamination reaction of the amine group onto another ketoacid or by deamination [81]. Myc, which is activated in most cancer types, increases glutaminolytic activity by increasing expression of *GLS* and the glutamine transporter *ASCT2* (ASC amino acid transporter 2) [82]. If a particular tumor uses this strategy to keep the Krebs cycle filled, it renders the cancer cells addicted to glutamine. The filling of the Krebs cycle via glutaminolysis is then used for several cataplerotic reactions important for cancer cell lipid biosynthesis: (1) malate is converted to pyruvate by malic enzyme (ME) in the cytosol, thereby yielding NADPH (Figure 3A), which is a reducing equivalent used for fatty acid biosynthesis; (2) oxaloacetate (OAA) is converted back to malate and also exported to yield NADPH; and (3) citrate is used as a source of acetyl CoA for fatty acid synthesis [83] (Figure 3A). The relative contribution of these reactions via ME to the NADPH pool is quite significant and is roughly equivalent to the NADPH production by the PPP [84].

Although glutaminolysis can replenish Krebs cycle intermediates, it still requires functional mitochondria and oxygen, which are not always available in the tumor context. Cancer cells use reductive carboxylation in cases of defective mitochondria [85] and hypoxia [86,87] (Figure 3B). This pathway involves NADPH-dependent reductive carboxylation of α-KG (that derives from glutaminolysis, see above paragraph) by isocitrate dehydrogenase (IDH) into isocitrate, which is the reverse reaction of the conventional Krebs cycle. Isocitrate is subsequently isomerized to citrate, which is essential for lipid biosynthesis. Substrate availability also influences the direction of the reaction: that is, reductive carboxylation or oxidative decarboxylation of α-KG. A high NADPH/$NADP^+$ (nicotinamide adenine dinucleotide phosphate) ratio, low citrate levels, and high α-KG abundance increase the reductive carboxylation activity of cells [88–91].

Cancer cells need to upregulate their fatty acid biosynthesis upon neoplastic transformation [92,93] in order to meet the increased demand of membrane production due to increased proliferation. Citrate production is of importance for fatty acid biosynthesis because the citrate shuttle transports mitochondrial acetyl CoA across the membranes to the cytoplasm, where it is converted back to OAA and acetyl CoA by ATP citrate lyase [94] (Figure 3A and B). This is one of the reasons why anaplerotic reactions are crucial for supporting growth and proliferation of the tumor cells.

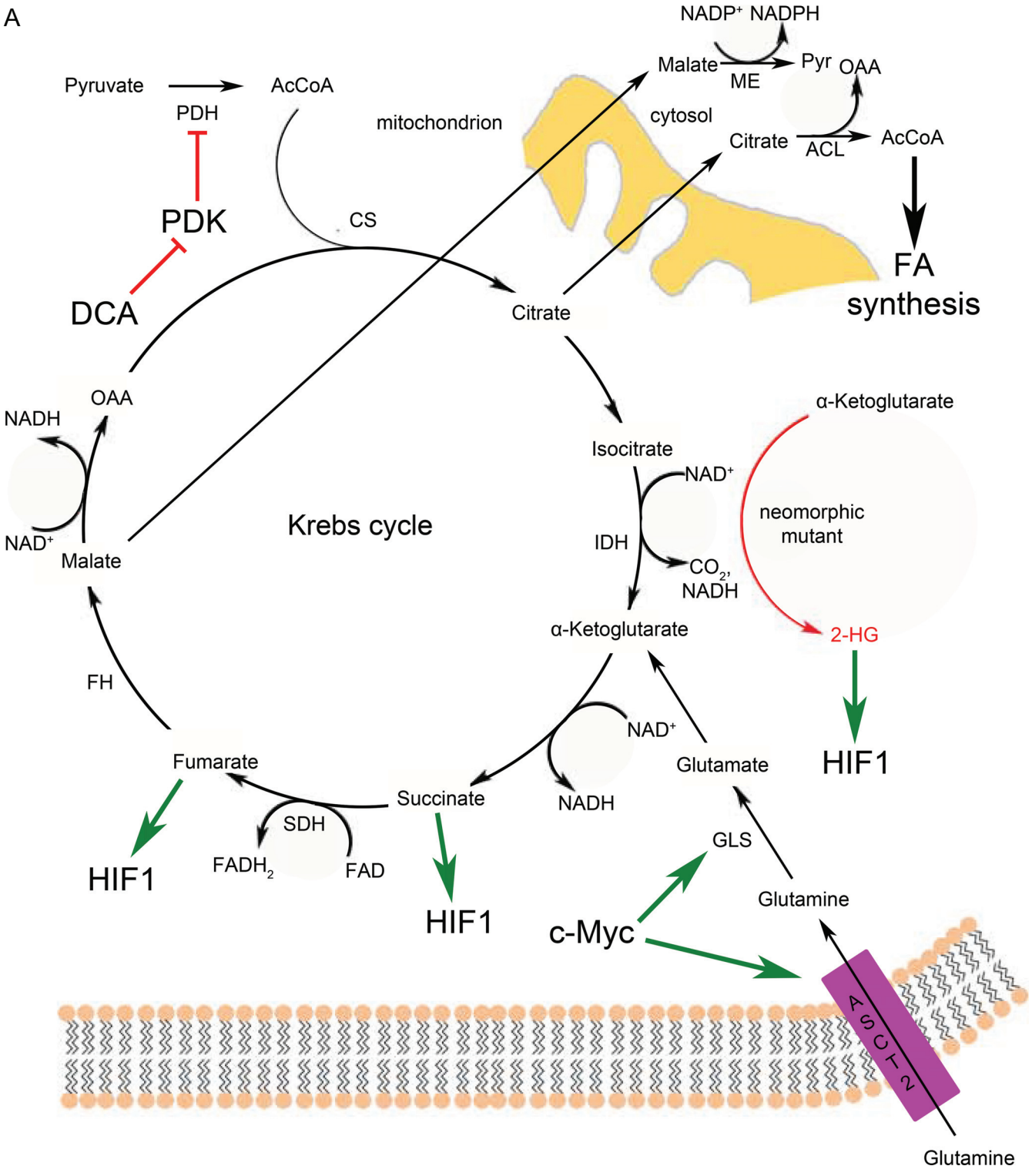
Warburg was not totally wrong

So far, we have discussed mechanisms that promote aerobic glycolysis independently of possible mitochondrial defects. However, there are cases where Warburg was right, whereby mutation of genes whose products act in mitochondria are enough to induce transformation. Discussed below are examples of such cases.

Various cancer types have been found to harbor neomorphic mutations in *IDH1* and *IDH2* enzymes, causing them to lose their activity of decarboxylation of isocitrate to α-KG, and to gain the activity of converting α-KG to the oncometabolite 2-hydroxyglutarate (2-HG) [95,96] (Figure 3A). The prevalent view on the mechanism of 2-HG-induced tumor formation is that it alters cellular methylation of both DNA and proteins. There is evidence that 2-HG competitively inhibits α-KG-dependent dioxygenases, such as methylcysteine hydroxylases and the Jumonji C (JmjC) family of histone demethylases [97–99]. Data also show that 2-HG stabilizes HIF-1α by inhibiting prolyl hydroxylases (PHDs) (also α-KG-dependent dioxygenases) [98].

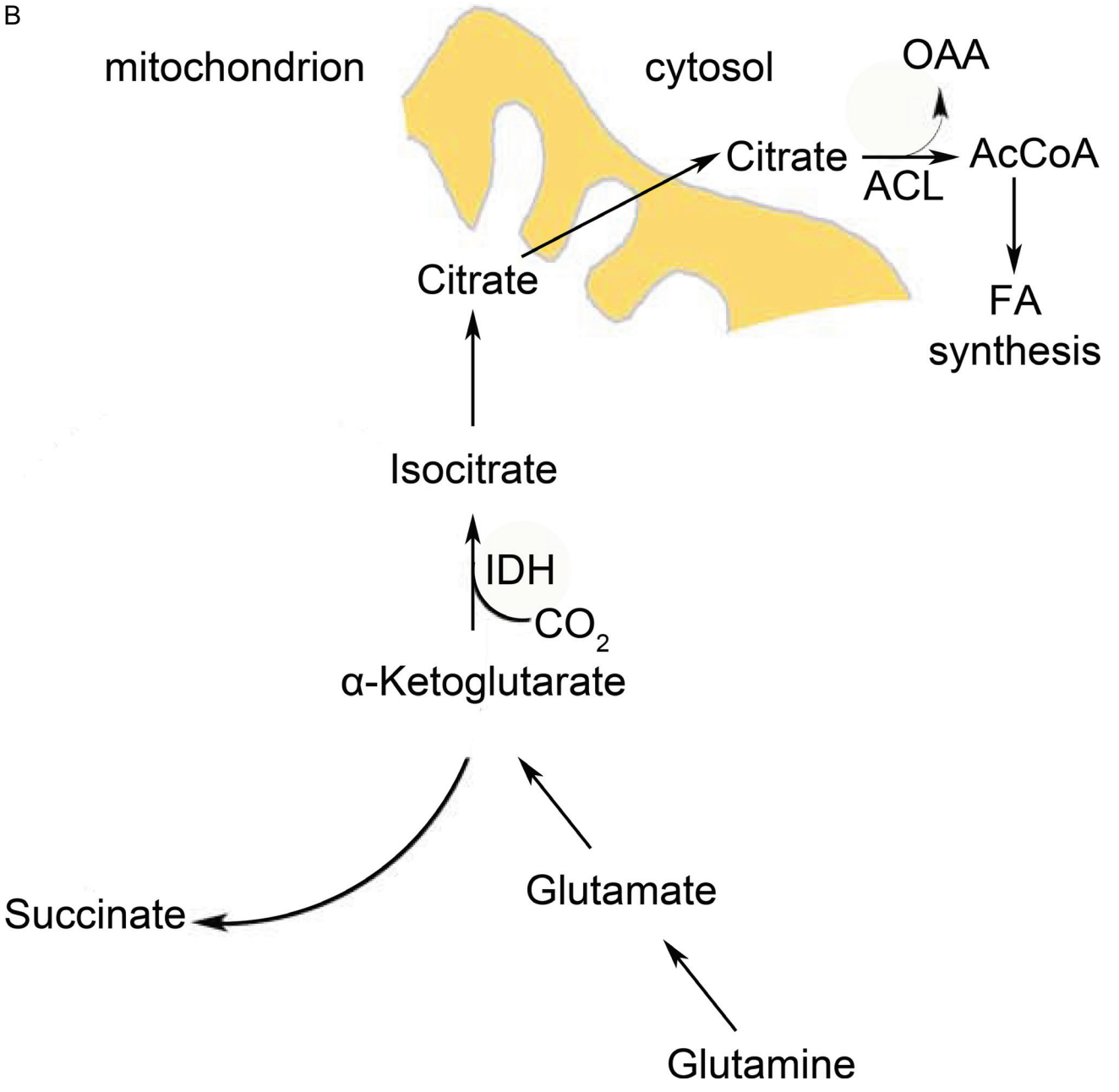
Some enzymes in the Krebs cycle function as tumor suppressors. For example, mutations in succinate dehydrogenase (SDH) subunits have been associated with hereditary paraganglioma [100,101], and fumarate hydratase (*FH*) mutations were implicated in renal cell cancers and smooth muscle tumors [102]. In both cases, substrates of the enzymes, succinate and fumarate, respectively, have been shown to accumulate. The mechanism of SDH and FH stemmed carcinogenesis is induction of a 'pseudohypoxia' state. Hypoxia-responsive genes are regulated by the transcription factor HIF1α. Under normoxic conditions, HIF1α is constantly hydroxylated at proline residues by PHDs. Upon hydroxylation, HIF1α is readily recognized and ubiquitinated by Von Hippel-Lindau complex that has E3 ubiquitin ligase activity [103,104]. PHDs use α-KG as cofactors and generate succinate at the end of the reaction [105]. In the context of SHD and *FH* mutations, cytosolic succinate [106,107] and fumarate [108] concentrations increase and they inhibit the activity of PHDs, leading to stabilization of HIF1α (Figure 3A) even in the presence of oxygen and induction of an aberrant hypoxic response [109].

Figure 3. Krebs cycle reactions are altered but not completely abrogated in cancer cells



(A) Full Krebs cycle in the presence of functional mitochondria and oxygen. Cancer cells use glutamine more than pyruvate for anaplerosis.

Figure 3. *Continued*



(B) A truncated form of the Krebs cycle is favored in cancer cells with defective mitochondria or under hypoxic conditions (or both) to generate metabolic precursors with reductive carboxylation. 2-HG, 2-hydroxyglutarate; AcCoA, acetyl coenzyme A; ACL, ATP citrate lyase; ASCT2, sodium-dependent neutral amino acid transporter type 2; CS, citrate synthase; DCA, dichloroacetate; FA, fatty acid; FAD, flavin adenine dinucleotide; FH, fumarate hydratase; GLS, glutaminase; HIF1, hypoxia-induced factor 1; IDH, isocitrate dehydrogenase; ME, malic enzyme; NAD, nicotinamide adenine dinucleotide; NADH, reduced nicotinamide adenine dinucleotide; NADP, nicotinamide adenine dinucleotide phosphate; NADPH, reduced nicotinamide adenine dinucleotide phosphate; OAA, oxaloacetate; PDH, pyruvate dehydrogenase; PDK, pyruvate dehydrogenase kinase; pyr, pyruvate; SDH, succinate dehydrogenase.

Conclusions

There has been discussion for almost a century about the reasons for the metabolic phenotypes that Warburg initially observed, i.e. that cells use glucose via glycolytic reactions instead of respiration, even in the presence of oxygen. It is a phenomenon that appears counter-intuitive and wasteful, given how little ATP glycolysis yields compared with oxidative phosphorylation. The

main disagreement revolves around Warburg's proposal that irreversible damage of mitochondria is the main reason for carcinogenesis. In some cases, Warburg's proposal holds true. For example, the fact that mutations in some Krebs cycle enzymes, such as SDH and FH, are able to induce renal tumors indicates that mitochondrial dysfunction can be sufficient for tumor formation. However, there are reports that in some cases cancer cell mitochondrial function is intact. When they are forced to respire, they are able to. In sum, it seems that mitochondrial dysfunction is usually the consequence of tumor formation, yet there are cases in which mitochondrial dysfunction is the cause of the tumor formation. Indeed, one recent study suggests that glycolysis is needed for sustaining stemness and proliferation, whereas forced activation of mitochondrial respiration induces differentiation and reduces proliferation [110]. There seems to be a very strong, yet not very well-understood, crosstalk between the regulation of proliferation and a preference for glycolysis.

Abbreviations

2-HG, 2-hydroxyglutarate; α-KG, α-ketoglutarate; AMPK, AMP-activated protein kinase; ATP, adenosine triphosphate; CoA, coenzyme A; DCA, dicholoroacetate; FDG, 2-fluoro-6-deoxyglucose; FH, fumarate hydratase; GLS, glutaminase; GLUT, glucose transporter; HIF, hypoxia-induced factor; HK, hexokinase; IDH, isocitrate dehydrogenase; LDH, lactate dehydrogenase; MCT4, monocarboxylate transporter 4; ME, malic enzyme; MPC, mitochondrial pyruvate carrier; NADH, reduced nicotinamide adenine dinucleotide; NADPH, reduced nicotinamide adenine dinucleotide phosphate; OAA, oxaloacetate; PDH, pyruvate dehydrogenase; PDK, pyruvate dehydrogenase kinase; PEP, phosphoenolpyruvate; PET, positron emission tomography; PFK, phosphofructokinase; PFKBP, phosphofructokinase/fructosebisphosphatase; PGAM, phosphoglycerate mutase; PGI, phosphoglucose isomerase; PHD, prolyl hydroxylase; PK, pyruvate kinase; PPP, pentose phosphate pathway; ROS, reactive oxygen species; SDH, succinate dehydrogenase; siRNA, small interfering RNA; TIGAR, TP53-induced glycolysis and apoptosis regulator; VDAC, voltage-dependent anion channel.

Disclosures

The authors declare that they have no disclosures.

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