Journal of Oncological Sciences 3 (2017) 45–51

Contents lists available at ScienceDirect

Journal of Oncological Sciences

journal homepage: https://www.elsevier.com/locate/jons

Biology of glucose metabolization in cancer cells

Adewale Fadaka*, Basiru Ajiboye, Oluwafemi Ojo, Olusola Adewale, Israel Olayide, Rosemary Emuowhochere

Department of Biochemistry, College of Science, Afe Babalola University, Ado-Ekiti, Ekiti State, Nigeria

ARTICLE INFO

Article history:
Received 12 April 2017
Received in revised form
3 June 2017
Accepted 14 June 2017
Available online 19 July 2017

Keywords:
Cancer
Glucose
Glycolysis
Hypoxia
Warburg

ABSTRACT

Cancer is a disease at the cellular level involving heritable disorders in cellular control mechanism. Cancer cells also need to adapt their metabolism to survive and multiply under the metabolically compromised conditions provided by the tumor microenvironment. Tumor cells alter their metabolism to maintain unregulated cellular proliferation and survival, but this transformation leaves them reliant on constant supply of nutrients and energy. They alter their metabolism to support their rapid proliferation and expansion across the body. After the discovery of based on the altered cancer cell metabolism in 1930, loads of studies have shed light on several aspects of cancer metabolism with a common goal to find new ways for effectively eliminating tumor cells by targeting their energy metabolism. Research has directed most of its resources to elucidate the causes, prevention and possible cure for cancer, yet the process has been elusive claiming human lives more than ever. This disease is a manifestation of etiological and pathological disturbances of mechanisms that control cell division, differentiation and metabolism. 50% of all human tumors carry genetic alterations that lead to the inactivation of some tumor suppressor proteins. Cancer cells are shown to experience characteristic changes in their metabolic programs, including increased uptake of glucose, enhanced rates of glutaminolysis and fatty acids synthesis, suggesting that metabolic shifts supports tumor cells growth and survival. In this review, we summarized the major concepts of glucose metabolization and explore the molecular basis of aerobic glycolysis of cancer cells.

1. Introduction

There has been an enigmatic search of information on the mechanism of cancer related metabolic adaptations, and this has resulted in accumulated evidences suggesting considerable association between several pathways in human metabolism and malignant transformation.[1–3] Carcinogenesis is a complex, multistep process requiring the elimination of several cell-imposed barriers such as anti-proliferative responses, programmed cell death-inducing mechanisms, and senescence. This occurs mostly through genetic alterations in oncogenes and tumor suppressor genes.[4] In other for the body to prevent the growth of tumor, there is chronic activation of the immune system. These two processes (tumor growth and immune system activation) are responsible for an increased energy usage and thus for continuous consumption of energetic substrates, such as carbohydrate.[5] In fact, the oxidation of glucose into CO_2 and H_2O through the citric acid cycle is a well-known major source of energy and plays a key role in the biosynthesis of ATP, constituent of DNA, RNA and phospholipids. Glucose is also necessary for the pentose phosphate pathway and the synthesis of reducing compounds such as NADPH. Energy metabolism in advanced tumor cells is severely compromised by the occurrence during the disease progression of symptoms such as nausea anorexia, and vomiting, which does not allow normal nutrition and therefore, a regular supply of carbohydrates, proteins, amino acids and vitamins. Reduced oral intake resulting from anorexia or obstruction of the gastrointestinal tract plays a crucial role in the development of the cancer cachexia syndrome. In addition to the reduced food intake, important changes of energy metabolism and biochemical/metabolic abnormalities in carbohydrate, protein and lipid biochemistry and metabolism have been observed, which may account for cancer-related anorexia/cachexia syndrome.[6] Altered energy metabolism consisting of increased resting energy expenditure associated with increased metabolism of sugar, lipid and

* Corresponding author.
E-mail addresses: fadakaao@abuad.edu.ng, silvernonferous@gmail.com (A. Fadaka).

Peer review under responsibility of Turkish Society of Medical Oncology.

http://dx.doi.org/10.1016/j.jons.2017.06.002

protein metabolism are typical of cancer cells. These changes are a consequence of alterations in intermediary metabolism (carbohydrate, protein, and lipids) associated with cancer. Also, activation of oncogenes and deactivation of tumor suppressor genes, have been associated to cancer-associated metabolic remodeling of epigenetic marks.[7]

2. Overview of carbohydrate metabolism in normal cell

Carbohydrates constitute a major part of our diet and our food is the ultimate source of all the sugars that enter our metabolic pathways. About two-thirds of dietary carbohydrate is the plant polysaccharide. Disaccharides such as lactose and sucrose and some other polysaccharides like cellulose are also a part of our food, but our intake of free monosaccharide like glucose, fructose and galactose is relatively reduced.[8]

Carbohydrate metabolism starts with digestion in the small intestine when monosaccharides are absorbed into the blood stream. Blood sugar concentrations are controlled by three hormones: insulin, glucagon, and epinephrine. If the concentration of glucose in the blood is increased insulin is released from the pancreas. Insulin stimulates the transfer of glucose into the cells in the liver and muscles, in the liver and muscles most of the glucose is converted to glycogen, a process known as glycogenesis. Glycogen is stored in the liver and muscle. The breakdown of glycogen to glucose is called glycogenolysis. The metabolites from glycolysis form links with protein, lipid and nucleic acid. Glucose is the central molecule in carbohydrate breakdown and synthesis. All major pathways of carbohydrate metabolism are connected to conversions of glucose, since glucose is the main sugar in the blood and the main energy fuel in the body. The metabolic pathways are glycolysis, Oxidation of Pyruvate, Citric Acid Cycle, Pentose Phosphate Pathway, Glycogen metabolism (glycogenolysis, glycogenesis) and Gluconeogenesis.

2.1. Glycolysis (Embden-Meyerhoff pathway) in normal cells

Glycolysis is a universal pathway for catabolism of glucose in animals and plants. It occurs in all cells of our body. The process (glycolysis) convert one molecule of glucose to two molecules of pyruvic acid, and the energy released is conserved in the form of ATP and reducing equivalent NADH. The main function or aim of glycolysis is to provide energy and intermediates for other metabolic pathways. The major sources of glucose for glycolysis are dietary carbohydrates and cellular glycogen. Glucose undergoes glycolysis by a sequence of 10 cytosolic reactions, seven of which are reversible while three are irreversible. The first five reactions constitute the “investment” phase since they use ATP, while the last five reactions constitute the “pay-off” phase and yield ATP. In glycolysis (normal cells), which happens in the cytoplasm, one glucose molecule yields 2 ATPs and 2 NADH molecules (NADH is another energy-carrying molecule), and two pyruvates. If oxygen is present, the two pyruvates, with help from the pyruvate dehydrogenase enzyme complex, is converted to two Acetyl-CoA molecules. The acetyl-CoAs enter the mitochondrion where it fuels the citric acid cycle. Each acetyl-CoA molecule goes through the citric acid cycle. Therefore, from one glucose, the yield is 6 NADH molecules, two $FADH_2$ molecules, and two ATP molecules. The 6 NADH +2 FADH, plus the NADH produced in glycolysis and at the PDH complex, now enter the “electron transport system” (ETS). This is where the efficient production of ATP occurs: The ETS is a multistage process, called oxidative phosphorylation or cellular respiration, which, with the help of ATP synthase has the following yield: This summary gives a theoretical yield of 40 ATP per glucose. 32–38 ATPs are generated depending on the enzymes an individual's DNA codes for. Cancer cells typically “switch” from “cellular respiration” to the very inefficient glycolysis for their ATP needs, a phenomenon described by Otto Warburg in 1924. The amazing thing is that tumors, which are highly energy demanding tissues, switch to a very inefficient energy producing pathway. They make up for this energy demand by going through glycolysis faster than necessary in normal cells.

2.2. Metabolizing pathways in cancer

Cancer is a disease condition arising from uncontrolled division of cells in the body to form lumps of tissues called tumors. There are many different types of cell in the body, and many different types of cancer which arise from different types of cell. Some are more easily treated than others, particularly if diagnosed at an early stage, some have a better outlook (prognosis) than others.[9] These cells may invade or spread to other parts of the body where they can go on proliferating and causing unhealthy condition to the system. Those cells with minimum uncontrolled growth and do not invade other cells are said to be benign while cells that grow and divide rapidly and as well invade other tissues are said to be malignant.

Normally, cells are expected to grow, divide and then die. In a situation where the deoxyribonucleic acid (DNA) of a cell is damaged by some factors, known as risk factors, and the cell cannot repair the damaged DNA, then it may not be able to control the normal programmed cell growth. This results to development of cancer. In each case, it is important to know exactly what type of cancer has developed, how large it has become, whether it has spread, and how well it usually responds to treatment. Cancer results from the development of abnormal properties in normal cells that enable them to grow excessively and spread to other locations. This abnormal development can be caused by mutations that occur from factors such as paints, chemicals, radiation, ultraviolet light and chromosome replication errors.[10] These mutagens alter DNA by changing nucleotide bases and can even change the shape of DNA. The altered DNA produces errors in DNA replication, as well as errors in protein synthesis. These changes influence cell growth, cell division, and aging. Viruses such as Human papilloma virus can cause cancer by altering cell genes though a class of cancer. Cancer viruses change cells by integrating their genetic material with the host cell's DNA.

2.2.1. The Crabtree effect

Crabtree made an observation on the utility of carbohydrates by cancer cells.[11] It was observed that, for normal cells, the presence of glucose slightly increased respiration or had no effect on oxygen consumption. On the contrary, glucose decreased oxygen uptake by cancer cells. This respiratory inhibition is known the Crabtree effect. It is now known that this metabolic transformation of cancer cells is not a specific feature of carcinogenesis, but appears to be a requirement of rapidly dividing cells such as proliferating hematopoietic progenitor cells, spermatozoa, intestinal mucosal cells, renal cells, and embryonic stem (ES) cells.[12] This phenomenon is also reported in bacteria and yeast. Apart from rapid proliferation, an important characteristic shared by all these cells, as will be expected from respiratory impairment, is increased glycolysis. Another observation of the Crabtree effect is the initial increase in respiration following the provision of glucose. Indeed, it appears that other hexoses can induce this effect in cancer cells as well. Several explanations have been provided based of the Crabtree effect, although the molecular mechanism is not fully understood. The mechanisms are as follows: (1) Competition for available ADP + Pi between oxidative phosphorylation and glycolysis can cause respiratory abnormalities by increased glucose availability.[13]

(2) Increased lactate production with decreasing cytosolic pH and inhibition of oxidative enzymes. (3) Disruption of coupled respiration by calcium; increased mitochondrial calcium levels by glucose caused an increased association of the inhibitory subunits of F_1F_0 to the ATP synthase to inhibit coupled respiration.[12] (4) Glucose metabolism increases reactive oxygen species production that damages mitochondrial membranes and depresses respiration (Fig. 2).[14] (5) The Crabtree effect might be regulated by several mechanisms in cancer cells, involving changes in ATP/ADP ratio, Pi, glucose-6-phosphate, cytosolic pH among others.[15] Irrespective of the mechanism, cancer cells are glycolytic and this is associated with partial mitochondrial impairment (i.e., suppressed respiration, less oxygen consumption, and low ATP production). Series of reactions that produce pyruvate is known as glycolysis. Under anaerobic conditions, glycolysis produces minimal energy in the form of ATP. This mode of glycolysis is not coupled to the TCA cycle and oxidative phosphorylation. However, under aerobic conditions, glycolysis, TCA cycle activity, and oxidative phosphorylation generate considerable energy for cellular functions. Glyceraldehyde 3-phosphate dehydrogenase (GAPDH) and the mitochondrial phosphate carrier (PiC) compete for the free cytoplasmic inorganic phosphate pool (Pi). A similar competition for free ADP occurs between the mitochondrial adenine nucleotide translocase (ANT) and two of the glycolysis enzymes: Phosphoglycerate kinase (PGK) and pyruvate kinase (PK). Fructose 1,6-biphosphate inhibits the mitochondrial respiratory complexes III and IV. Increased calcium (Ca_2^+) accumulation in the mitochondrial matrix lead to the inhibition of the ATP synthase.[16]

2.2.2. The Warburg phenomenon

While it can be conceived that fast-growing cancer cells will require more energy than normal cells, it is ironic to realize that, in contrast to normal cells, cancer cells use a primitive inefficient reaction, aerobic glycolysis, to generate considerable amounts of their energy. A possible reason for this bioenergetic alteration is the requirement to produce other metabolic end products to support their rapid growth and proliferation under low oxygen tension, and the possible adaptation to evade death in toxic environments or because of cytotoxic agents. Probably the most important treatise ever provided for mitochondrial dysfunction and its possible causative role of cancer is that provided several decades ago by the Nobel Laureate and Biochemist, Otto Warburg. In his series of experiments on respiration and metabolism of cancer cells, coupled with his in-depth analysis of reported works from other investigators at the time, by an approach reminiscent of what Watson and Crick employed in deciphering the DNA double helical structure, Otto Warburg could unwaveringly hypothesize that neoplastic transformation originated because of irreversible damage to mitochondrial respiration. Cancer cells are, therefore, compelled to rely on the inefficient glycolytic mode of ATP synthesis (2 ATPs/glucose), rather than respiration that produces substantially more ATP/glucose (approximately 36 ATPs/glucose). Warburg observed that, in conditions of normal oxygen tension, normal cells produced most of their energy via mitochondrial respiration. In contrast, over 50% of cancer cell energy was generated in the cytosol via glycolysis, with the remainder from the mitochondrial respiratory chain. This bioenergetically inefficient glycolytic reliance of cancer cells for most of their energy production is not primarily due to lack of oxygen, because it operates even in the presence of adequate oxygenation. The bioenergetically inferior nature of glycolysis implies that cancer cells must adopt a mode of increased glucose import to meet their energy demands. The mechanisms of aerobic glycolysis (Warburg effect) of cancer cells. Glucose transporters (Glut1 and Glut3) in the plasma membrane are overexpressed. Hexokinase isoform II (HK II) interacts with mitochondria through the voltage-dependent anionic channel (VDAC). Phosphofructokinase (PFK) is overactive because the predominance of its L- and P-isoforms. An overexpressed PFKB3 maintains higher levels of fructose 2,6-biphosphate that further activate PFK. The M2 isoform of pyruvate kinase (PK) regulates the glycolytic flux and promotes metabolite accumulation to fulfill the biosynthetic needs of the cell. The M-isoform of lactate dehydrogenase (LDH) is overexpressed. Pyruvate transport into the mitochondrial matrix through its specific carrier (PC) is decreased. Pyruvate dehydrogenase complex is inhibited in a phosphorylation-dependent mechanism mediated by an over-expressed pyruvate dehydrogenase kinase (PDHK). Mutations in succinate dehydrogenase (SDH) impair its activity. The levels of the mitochondrial respiratory complexes I, III and IV are decreased. Mitochondria ATP synthase activity is restricted by the overexpression of its inhibitory subunit.[16]

2.2.3. Differences between cancer cells and normal cells

Cancer cells differ from normal cells in many ways that allow them to grow out of control and become invasive. One important difference is that cancer cells are less specialized than normal cells.[17] This is one reason that, unlike normal cells, cancer cells continue to divide uncontrollably. In addition, cancer cells can ignore signals that normally tell cells to stop dividing or that begin a process known as programmed cell death, or apoptosis, which the body uses to get rid of unneeded cells by producing large quantity of CD47. Cancer cells may be able to influence the normal cells, molecules and blood vessels that surround and feed a tumor (an area known as the micro-environment).[10] For example, cancer cells can induce nearby normal cells to form blood vessels that supply tumors with oxygen and nutrients, which they need to grow (Fig. 1). Cancer cells are also often able to evade the immune system, a network of organs, tissues, and specialized cells that protects the body from infections and other conditions. Although the immune system normally removes damaged or abnormal cells from the body, some cancer cells can “hide” from the immune system.[18] Cancer cells exhibit aerobic glycolysis. This means that cancer cells derive most of their energy from glycolysis that is glucose is converted to lactate for energy followed by lactate fermentation, even when oxygen is available. This is termed the Warburg effect. This produces far less energy than oxidative phosphorylation and as cancer cells require a lot of energy to grow this seems paradoxical. The Warburg effect may be beneficial to cancer cells however because it provides precursors for many biosynthetic pathways. These precursors include amino acid precursors and NADPH and ribose sugars for DNA and RNA synthesis. The Warburg effect may be caused by impaired oxygen sensing in cancer cells or by the Myc transcription factor which is unregulated in cancer cells. Myc bypasses the normal oxygen sensing mechanism and results in activation of transcription of glycolytic enzymes. Glycolytic enzymes such as GLUT1, lactate dehydrogenase, pyruvate kinase and the lactate exporter are unregulated in cancer cells whilst pyruvate dehydrogenase is inhibited leading to increased glycolytic flux and impaired ability of pyruvate to enter oxidative phosphorylation.

3. The effects of hypoxia on glucose utilization in tumors

The availability of growth regulating factors control normal cell proliferation in tissues and by the interaction with surrounding cells. Nutrients and oxygen availability, necessary for cell proliferation and metabolism, largely depends on blood supply. Initial tumor growth occurs in the absence of formation of new blood vessels. Tumor cells bypass environmental growth-controlling constraints (Fig. 2). This is achieved by acquiring the ability to proliferate independently of growth signals, though, for example, mutations in receptor-associated signaling molecules, and by

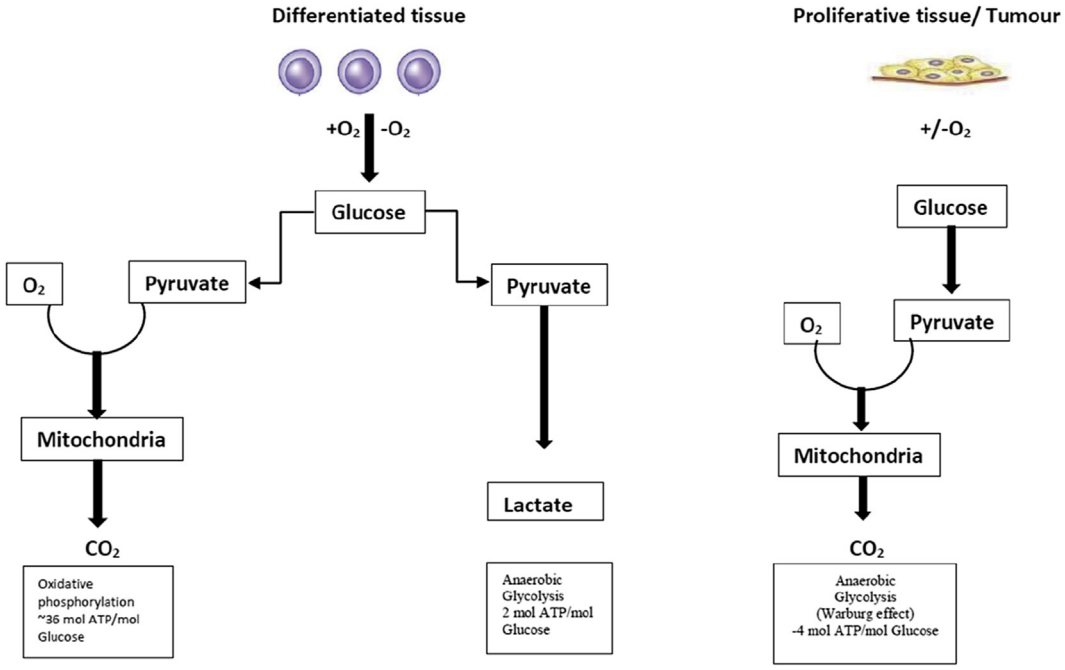

Fig. 1. Schematic diagram of aerobic glycolysis in cancer cell compared with normal cell.

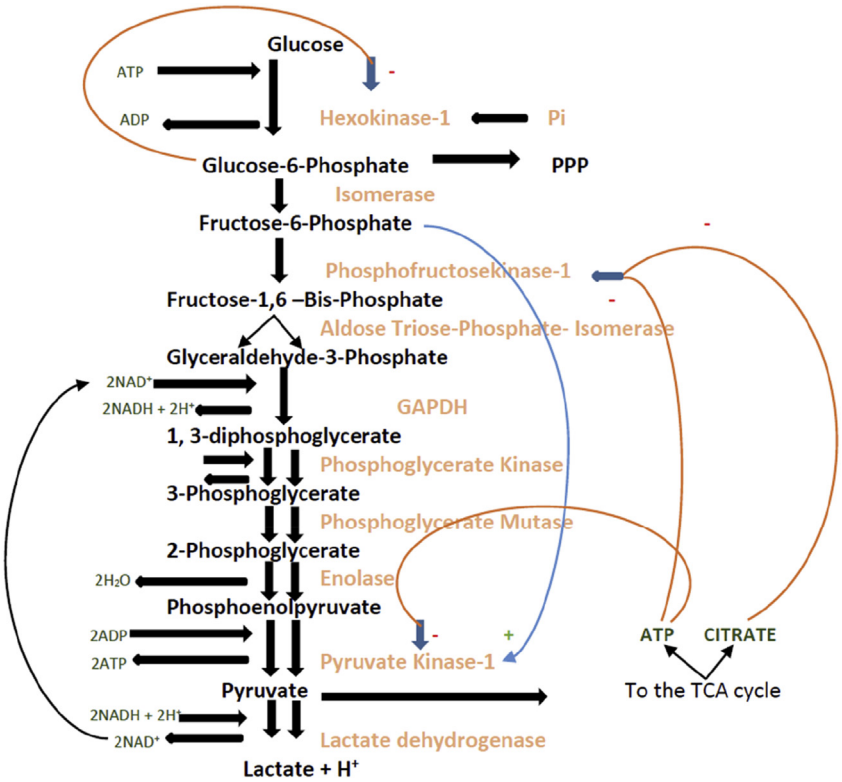

Fig. 2. Glycolysis in tumor cell. (Allosteric regulations of glycolysis confer metabolic plasticity with respect to local pO_2).

becoming insensitive to antigrowth stimuli, such as those mediated by cell-to-cell contacts. In the early carcinogenesis phase, uncontrolled cell proliferation moves tumor cells away from blood vessels and, therefore, from oxygen and nutrient supply. The only way oxygen and glucose can reach the inner cells of a non-vascularized tumor is by diffusion across the basement membrane and through the peripheral tumor-cell layers. However, partial oxygen pressure drops to very low values 100 mm away from blood vessels.

3.1. Tumor fuel metabolism

The main metabolic pathways that contribute to malignancy and offer potential drug targets are illustrated. Metabolic enzymes that have been associated with tumor initiation and growth are

marked in red. GLUT1, glucose transporter; HK, hexokinase; 6PGD, 6-Phosphogluconate dehydrogenase; PFKFB 3,6-phosphofructo-2-kinase; GAPDH, Glyceraldehyde3-phosphatedehydrogenase; PHGDH, phosphoglycerate dehydrogenase; PGAM1, Phosphoglyceratemutase1; PKM, Pyruvate kinase; LDHA, lactated hydrogenase A; MCT, Monocarboxylate transporter; PDH, Pyruvate dehydrogenase; CPT1, Carnitine palmitoyl transferase I; FASN, fatty acid synthase; RNR, ribonucleotide reductase; FH, fumarate hydratase; SDH, succinate dehydrogenase; IDH, isocitrate dehydrogenase; GLUD, glutamate dehydrogenase; GLS1, glutaminase1; ASCT2, Amino-acid transporter2; ACLY, ATP citrate lyase; ACC, acetyl-CoAcarboxylase; ACSS2, Acetyl CoA synthetase 2; DHFR, DHF reductase; TYMS, thymidylate synthase; HMGCR, HMG-CoA reductase; CK, choline kinase; G6P, glucose-6-phosphate; F6P, fructose-6-phosphate; PRPP, 5-phospho-alpha-D-ribose1-diphosphate; IMP, inosine monophosphate; UMP, uridine monophosphate; dNTP, deoxynucleotide triphosphate; G3P, glyceraldehyde3-phosphate; 3PG, 3-phosphoglycerate; 2PG, 2-phosphoglycerate; PEP, phosphoenol pyruvate; OAA, Oxaloacetate; AKG, a-ketoglutarate; HMG-CoA, 3-hydroxy-3-methyl-glutaryl coenzymeA; THF, tetrahydrofolate; 5, 10mTHF, 5, 10-methylenetetrahydrofolate; DHF, dihydrofolate.[19] Hypoxia and glucose shortage are rapidly generated in the inner mass of a growing tumor. Paradoxically, however, it is known since the 1920s.[20] that tumor cells have a much higher rate of glucose consumption through a glycolysis pathway that prevent pyruvate from entering the Krebs cycle and converts pyruvate to lactate: the so-called Warburg effect.[1] In fact, many tumors use this glucose to lactate pathway even in the presence of oxygen, and therefore, the term aerobic glycolysis is often used as a substitute to the Warburg effect. It is important to note that the glycolytic switch occurring in cancer cells is not necessarily accompanied by a reduction in oxidative phosphorylation.[21] Nowadays, the augmented glycolytic activity of tumors is clinically exploited by positron emission tomography for the identification of metastatic lesions. This technique takes advantage of the increased ability of tumor cells to take up and metabolize glucose compared with normal tissues.[10] It can therefore be stated that hypoxia is what drives tumor cells to fuel glucose in a non-oxidative 'glucose to lactate' pathway. However, it is currently believed that the glycolytic switch is acquired very early in carcinogenesis even before tumors experience hypoxia.[1] Consequently, the fact that, even in normoxic conditions, many tumors use aerobic glycolysis for their metabolic requirements indicates that the Warburg effect has functions that are not solely limited to hypoxia adaptation.

3.2. Cancer cell adaptation to hypoxic conditions

The transcription factor hypoxia-inducible factor-1 (HIF-1) is a key regulator responsible for the induction of genes that facilitate adaptation and survival of cells and the whole organism from normoxia ($\sim$ 21% O_2) to hypoxia ($\sim$ 1% O_2).[22,23] Since the identification of HIF, knowledge has grown exponentially. Because of the realization that hypoxia has a strong impact, via gene expression, on cell biology and mammalian physiology. HIF-1 is a pleiotropic transcription factor that regulates genes involved in the hypoxia induced metabolic switch, regulation of tumor pH, and angiogenesis.[24] The high glycolytic rate characteristic of hypoxic solid tumor is due in part to the greatly increased expression of hexokinase II (HK II),[25] a known transcriptional target of HIF-1. Hexokinase catalyses the first step in the glycolytic pathway where glucose is phosphorylated to glucose-6-phosphate with conversion of one ATP to ADP. There are four isoforms encoded by the mammalian genomes that are usually expressed at low levels in cells.[26] By increasing the expression level of HK II, hypoxia via HIF-1, can therefore modulate glucose metabolism. HK II has additional features that are relevant in the context of cancer-cell apoptosis. HK II is normally associated with voltage-dependent anion channel (VDAC).[27] A 30kDa pore protein inserted in the outer mitochondrial membrane that regulates the transport of metabolites in and out of the mitochondrial inter-membrane space.[28] Mitochondria are key components of the apoptotic cell death process. Upon exposure to cell-death stimuli, mitochondria release cytochrome-c and other apoptogenic factors into the cytoplasm where they trigger caspase activation and apoptosis.[29] Although the exact mechanism by which this happens remains unclear. One model proposes that Bax, once activated by death stimuli, cooperates with VDAC to form a large cytochrome-c conducting channel through the mitochondrial membrane.[30] It can be envisioned that a protein interacting with VDAC, like HK II, has the potential to prevent the interaction of proapoptotic proteins with mitochondria and consequently interfere with apoptosis. Therefore, in hypoxic tumors, the initial overexpression of HK II as a primary adaptation to hypoxia may secondarily confer resistance to apoptosis. Supporting this notion is the observation that disruption of the binding of HK II to mitochondria, through activation of GSK3b and phosphorylation of VDAC, potentiates chemotherapy induced cytotoxicity in transformed cells.[31] Moreover, methyl jasmonate, an anticancer agent that interacts directly with mitochondria can induce apoptosis selectively in cancer cells apparently by detaching hexokinases from mitochondria.[32] Recently, it has also been shown that the release of HK II from mitochondria potentiates cisplatin-induced cytotoxicity.[33] Current evidence indicates that hypoxia response in cancer cells, in addition to modulating glucose metabolism, renders them more resistant to death stimuli. Hypoxic tumors remain invasive and metastatic.[34] The best-characterized regulatory mechanism is that modulating HIF-1α's stability. Under well-oxygenated normoxic conditions, prolyl hydroxylation (by prolyl hydroxylases (PHDs)) and subsequent ubiquitination (by von-Hippel Lindau (VHL)-containing E3 ubiquitin-protein ligase) of the oxygen-dependent degradation (ODD) domain of HIF-1α leads to rapid degradation of HIF-1α with a half-life of 5–8 min. Consequently, HIF-1 is inactive under normoxic conditions.[35–37] On the other hand, under oxygen-deprived hypoxic conditions, HIF-1α becomes stable because oxygen depletion directly decreases the PHDs' activity.[36] Then, HIF-1α interacts with HIF-1β, forms a heterodimer, HIF-1,[22] binds to its cognate DNA sequence, the hypoxic-responsive element (HRE), and finally induces the expression of various genes related to angiogenesis, metastasis, glycolysis.[38–40] In addition to the regulation of HIF-1α's stability, another post-translational modification of HIF-1α is known to function in the regulation of the transactivational activity of HIF-1. Under normoxic conditions, factor inhibiting HIF-1 (FIH-1) becomes active and hydroxylates an asparagine residue of HIF-1α.[35,37] The asparaginyl hydroxylation blocks the interaction of HIF-1α with the transcriptional co-factor p300 and CBP, resulting in the suppression of HIF-1's transactivational activity. Because oxygen is a substrate of FIH-1 as well as PHDs, HIF-1's transactivational activity is restored under oxygen-deprived hypoxic conditions.[41]

3.3. The effect of tumor microenvironment on the tumor metabolism

Low oxygen concentration within solid tumors because of abnormal blood vessel formation, defective blood perfusion, and unlimited cancer cell proliferation is referred to as hypoxia. As tumor growth outpaces that of adequate vasculature, oxygen and nutrient delivery become insufficient. This dynamic interplay between the normal stroma and the malignant parenchyma, coupled with inevitable hypoxia, is common in any solid tumor

microenvironment. Interactions between malignant and non-transformed cells create the tumor microenvironment (TME). The progression of hypoxia over time is a consequence of increased oxygen consumption by abnormally proliferating cancer cells, which also produce an acidic environment. Most human tumors are highly heterogeneous and involve diverse microenvironments. A typical microenvironment seen in solid tumors is hypoxia, low-oxygen conditions under physiological level.[42] Tumor hypoxia is a concern in cancer therapy because it increases the metastatic and angiogenic potential of cancer cells,[38,43] and can render cancer cells resistant to radiation and chemotherapy.[44] Apart from malignant cells, the TME contains cells of the immune system, the tumor vasculature and lymphatics, as well as fibroblasts, pericytes and sometimes adipocytes, which are discussed in detail below. These cells are frequently distinguished by cell-type-specific markers, which are often cell surface molecules. The tumor microenvironment is a complex scaffold of an extracellular matrix (ECM) and various cell types. In addition to malignant cells, vascular cells, stromal cells and immune cells are common cellular residents of the tumor niche. Tumor cells mould this environment for their own needs via intercellular communication pathways, such as direct cell-to-cell contacts and the release of growth factors, matrix metalloproteases, ECM proteins and extracellular vesicles (EVs). Tumor cell-mediated stromal modifications include: suppression of anti-tumoural immune responses, deposition and degradation of ECM components, induction of vascular network formation and recruitment of stromal cells and tumor-promoting immune cells. In turn, heterogeneous tumor microenvironmental components create a favorable environment for tumor growth and dissemination. Various tumor microenvironmental stressors are inherent features of solid tumors that profoundly modify the tumor milieu and accelerate tumor progression towards malignancy.[45]

4. Conclusion

Cancer cells under hypoxic conditions have been recognized as crucial and excellent targets for cancer therapy because they mediate tumor malignancy and resistance to conventional treatments and seen in malignant tumors, not in normal tissues. Countless approaches have been used as targets; hypoxia-targeting using hypoxia-responsive promoters and hypoxia-specific replication of adenovirus as well as hypoxic cytotoxins and HIF-1 inhibitors. Hypoxia/HIF-1-targeting gene therapy is a promising tumor-specific approach with few side effects in normal tissues, and has the potential to enhance the effect of radiation therapy. Some approaches are now in clinical trials and are expected to lead to breakthroughs in cancer therapy. The elucidation of how a metabolism change with important genes and enzyme mutation causes cancer will liberate future novel cancer treatment development. Also, the manipulation of glucose metabolism with the immune system should also be subject of intense interest to ameliorate cancer progression.

References

1. Vander-Heiden MG, Cantley LC, Thompson CB. Understanding the Warburg effect: the metabolic requirements of cell proliferation. *Science*. 2009;324:1029–1033. http://dx.doi.org/10.1126/science.1160809.
2. Cairns RA, Harris IS, Mak TW. Regulation of cancer cell metabolism. *Nat Rev.Cancer*. 2011;11:8–95. http://dx.doi.org/10.1038/nrc2981.
3. Schulze A, Harris AL. How cancer metabolism is tuned for proliferation and vulnerable to disruption. *Nature*. 2012;491:364–373. http://dx.doi.org/10.1038/nature11706.
4. Bartkova J, Rezaei N, Liontos M. Oncogene-induced senescence is part of the tumor genesis barrier imposed by DNA damage checkpoints. *Nature*. 2006;444:633–637.
5. Mantovani G, Maccio A, Lai P. Cytokine activity in cancer-related anorexia/cachexia: role of megestrol acetate and medroxyprogesterone acetate. *Semin Oncol*. 1998;25(Suppl. 6):45–52.
6. Mantovani G, Maccio A, Massa E, Madeddu C. Managing cancer-related anorexia/cachexia. *Drugs*. 2001;61:499–514.
7. Levine AJ, Puzio-Kuter AM. The control of the metabolic switch in cancers by oncogenes and tumor suppressor genes. *Science*. 2010;330:1340–1344. http://dx.doi.org/10.1126/science.1193494.
8. Berg JM, Tymoczko JL, Stryer L. *Biochemistry*. sixth ed. New York: WH. Freeman; 2006.
9. Hatzivassiliou G. ATP citrate lyase inhibition can suppress tumor cell growth. *Cancer Cell*. 2005;8:311.
10. Gatenby RA, Gillies RJ. Why do cancers have high aerobic glycolysis? *Nat Cancer*. 2004;4:891–899.
11. Crabtree HG. Observations on the carbohydrate metabolism of tumours. *Biochem J*. 1926;23:536–545.
12. Wojtczak L. The Crabtree effect: a new look at the old problem. *Acta Biochim Pol*. 1996;43:361–368.
13. Sussman I, Erecinska M, Wilson DF. Regulation of cellular energy metabolism: the Crabtree effect. *Biochim Biophys Acta*. 1980;591:209–223.
14. Yang X, Borg LA, Eriksson UJ. Altered metabolism and superoxide generation in neural tissue of rat embryos exposed to high glucose. *Am J Physiol*. 1997;272:173–180.
15. Rodriguez-Enriquez S, Juarez O, Rodriguez-Zavala JS, Moreno-Sanchez R. Multisite control of the Crabtree effect in ascites hepatoma cells. *Eur J Biochem*. 2001;268:2512–2519.
16. Diaz-Ruiz R, Rigoulet M, Devin A. The Warburg and Crabtree effects: on the origin of cancer cell energy metabolism and of yeast glucose repression. *Biochimica Biophys. Acta*. 2011;1807:568–576.
17. Elstrom RL, Bauer DE, Buzzai M. Akt stimulates aerobic glycolysis in cancer cells. *Cancer Res*. 2004;64:3892–3899.
18. Bensaad K, Tsuruta A, Selak MA, Vidal MN, Nakano K, Bartrons R. TIGAR, a p53 inducible regulator of glycolysis and apoptosis. *Cell*. 2006;126:107–120.
19. Porporato PE, Dhup S, Dadhich RK, Copetti T, Sonveaux E. Anticancer targets in the glycolytic metabolism of tumors: a comprehensive review. Frontiers in Pharmacology. *Pharmacol Anti-Cancer Drugs*. 2011;2:4916.
20. Warburg O, Wind F, Negelein E. The metabolism of tumors in the body. *J Gen Physiol*. 1927;123:309–314.
21. Moreno-Sánchez R, Rodríguez-Enríquez S, Marín-Hernández A, Saavedra E. Energy metabolism in tumor cells. *FEBS J*. 2007;274:1393–1418.
22. Wang GL, Jiang BH, Rue EA, Semenza GL. Hypoxia-inducible factor 1 is a basic-helix-loop helix-PAS heterodimer regulated by cellular O_2 tension. *Proc Natl Acad Sci U. S. A*. 1995;92(12):5510–5514.
23. Semenza GL. Hypoxia-inducible factor 1: master regulator of O_2 homeostasis. *Curr Opin Genet Dev*. 1998;8:588–594.
24. Pouyssegur J, Dayan F, Mazure NM. Hypoxia signaling in cancer and approaches to enforce tumour regression. *Nature*. 2006;441:437–443.
25. Rempel A, Mathupala SP, Griffin CA. Glucose catabolism in cancer cells: amplification of the gene encoding type II hexokinase. *Cancer Res1*. 1996;56:2468–2471.
26. Wilson JE. Isozymes of mammalian hexokinase: structure, subcellular localization and metabolic function. *J Exp Biol*. 2003;206:2049–2057.
27. Mathupala SP, Ko YH, Pedersen PL. Hexokinase II: cancer's double-edges word acting as both facilitator and gatekeeper of malignancy when bound to mitochondria. *Oncogene*. 2006;25:4777–4786.
28. Colombini M. VDAC: the channel at the interface between mitochondria and the cytosol. *Mol Cell Biochem*. 2004;256–257:107–115.
29. Taylor RC, Cullen SP, Martin SJ. Apoptosis: controlled demolition at the cellular level. *Nat Rev Mol Cell Biol*. 2008;9:231–241.
30. Shimizu S, Matsuoka Y, Shinohara Y. Essential role of voltage-dependent anion channel in various forms of apoptosis in mammalian cells. *J Cell Biol*. 2001;152:237–250.
31. Pastorino JG, Hoek JB, Shulga N. Activation of glycogen synthase kinase 3b-disrupts the binding of hexokinase-II to mitochondria by phosphorylating voltage-dependent anion channel and potentiates chemotherapy-induced cytotoxicity. *Cancer Res*. 2005;65:10545–10554.
32. Goldin N, Arzoine L, Heyfets A. Methyl jasmonate binds to and detaches mitochondria-bound hexokinase. *Oncogene*. 2008;27:4636–4643.
33. Shulga N, Wilson-Smith R, Pastorino JG. Hexokinase II detachment from the mitochondria potentiates cisplatin induced cytotoxicity through a caspase-2dependent mechanism. *Cell Cycle*. 2009;8:3355–3364.
34. Cairns RA, Kalliomaki T, Hill RP. Acute (cyclic) hypoxia enhances spontaneous metastasis of KHT murine tumors. *Cancer Res*. 2001;61:8903–8908.
35. Hirota K, Semenza GL. Regulation of hypoxia-inducible factor 1 by prolyl and asparaginyl hydroxylases. *Biochem Biophys. Res Commun*. 2005;338:610–616.
36. Jaakkola P, Mole DR, Tian YM, Wilson MI, Gielbert J, Gaskell SJ. Targeting of HIF-alpha to the von Hippel Lindau ubiquitylation complex by O_2-regulated prolyl hydroxylation. *Science*. 2001;292(5516):468–472.
37. Semenza GL. HIF-1, O(2), and the 3 PHDs: how animal cells signal hypoxia to the nucleus. *Cell*. 2001;107(1):1–3.
38. Erler JT, Bennewith KL, Nicolau M, Dornhofer N, Kong C, Le QT. Lysyl oxidase is essential for hypoxia-induced metastasis. *Nature*. 2006;440(7088):1222–1226.
39. Kim JW, Gao P, Dang CV. Effects of hypoxia on tumour metabolism. *Cancer Metastasis Rev*. 2007;26(2):291–298.
40. Rofstad EK. Microenvironment-induced cancer metastasis. *Int J Radiat Biol*. 2000;76(5):589–605.
41. Harada H, 978-953-307-540-2. In: Prof YongpingYou, ed. *Gene Therapy Strategy*

for Tumour Hypoxia, Targets in Gene Therapy. InTech; 2011. Available from: http://www.intechopen.com/books/targetsingene therapy/gene-therapy-strategy-for-tumour hypoxia.
42. Thomlinson RH, Gray LH. The histological structure of some human lung cancers and the possible implications for radiotherapy. *Br J Cancer*. 1955;9(4):539–549.
43. Yang MH, Wu MZ, Chiou SH, Chen PM, Chang SY, Liu CJ. Direct regulation of TWISTby HIF-1alpha promotes metastasis. *Nat Cell Biol*. 2008;10(3):295–305.
44. Brown JM, Wilson WR. Exploiting tumour hypoxia in cancer treatment. *Nat Rev Cancer*. 2004;4(6):437–447.
45. Kucharzewska P, Belting M. Emerging roles of extracellular vesicles in the adaptive response of tumour cells to micro-environmental stress. *J. extracellular vesicles*. 2013;2. http://dx.doi.org/10.3402/jev.v2i0.20304, 20304.