MINI-REVIEW

Metabolism and Cancer: An Up-to-date Review of a Mutual Connection

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Abstract

Cancer and metabolism, namely that of glucose, share a reciprocal relationship. Discovered 80 years ago, the so-called Warburg effect remains a conundrum both teleologically and mechanistically and thus, has been thoughtfully endowed with respectful attention. With the progress of years, scientific contributions have managed to solve parts of this mystery. The present review aims to be a reviewing prelude to most recent findings regarding the connection of cancer with glucose metabolism.

Keywords: Cancer - metabolism - glucose - carcinogenesis - genes

Introduction

The spotlight of the apparently "friendly" relation that cancer cells exhibit with glucose metabolism commenced 80 years ago, when Warburg observed than in contrast to normal tissues, cancer cells tend to convert glucose into lactate even in the presence of sufficient oxygen, a term called Warburg effect or aerobic glycolysis. In other words, although all cells present the so-called "Pasteur effect", the shift to glycolysis in the absence of oxygen, it is cancer cells that present in addition the "Warburg effect". Proposed initially by Warburg to be a leading cancerous phenomenon, it has received much less attention in the following years of oncogenes' discovery as primary cancer players (Shaw, 2006), thus formulating a "chicken or egg" relation.

Although it is clear like water that abnormal uptake of nutrients is observed in cancerous cells, mostly due to relevant oncogenic mutations, what Warburg effect serves for remains obscure (Vander Heiden et al., 2009). Nevertheless, nowadays it has been shown that the Warburg hypothesis of defective mitochondrion is not taking place in the cellular environment (Fantin et al., 2006).

Teleologically, increased glycolysis in cancer cells, as extensively reviewed by Yalcin et al., (2009), contributes to: 1. Rapid production of ATP. 2. Synthesis of intermediates for biosynthetic pathways. 3. Creation of low pH levels both intra- and extra-cellularly. Mechanistically, the 6-phosphofructo-2-kinase/ fructose-2, 6-bisphosphatases (PFK2/FBPase), which are implicated in phosphorylating fructose-6-phosphate to fructose-2, 6-biphosphate and in the inverse dephosporylation, are regarded to contribute to the highly glycolytic rates in cancerous versus normal cells, as PFK2/FBPase exhibit the highest kinase: phosphatase ratio (Telang et al., 2009).

With regard to cancer, three oncogenes have mainly been related to promote the Warburg effect: Akt, Myc and Ras (Hsu and Sabatini, 2009). Akt controls regulation of phosphofructokinase-2 as well as translocation of hexokinase to the mitochondria (Shaw, 2006). Myc has been reported to increase oxidative metabolism of glucose during cell-cycle entry indicated by a remarkably higher activity of pyruvate dehydrogenase and of supply of acetyl-CoA by pyruvate in myc+/+ versus myc-/- cells (Morrish et al., 2009).

Nevertheless, in contrast to oxidative phosphoryation, even aerobic glycolysis is considered a less efficient metabolism. Two possible explanations for why have been presented in a thorough review by Vander Heiden et al., (2009): the one is that ineffective ATP production is problematic only when there is a scarcity of sources, while the second explanation is that cells which are proliferating have important requirements that extend beyond ATP. The same authors underline also that the explanation by which tumour hypoxia is a selective advantage for cells depending on anaerobic metabolism is under considerable doubts, since hypoxia seems not be a primary but a late-occuring phenomenon enough after the switch of cancer cells to aerobic glycolysis.

Notably, Lum et al. (2007) showed that hypoxia-inducible factor (HIF) plays a role for glucose metabolism even in the absence of hypoxia. Still, though, HIF-1 overexpression is in correlation with poor survival in different form of solid tumours, which present hypoxic regions. Interestingly, hypoxia-mediated HIF-1 expression leads to the expression of specific isoforms of glycolytic enzymes and transporters through alternative splicing (Marín-Hernández et al., 2009). The underlying mechanism remains elusive, athough it is understood that

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pre-mRNA splicing plays a core role in "metaphrasing" cellular stress to gene-expression profiles (Biamonti and Caceres, 2009).

Furthermore, the phylogenetical analogy of microbial fermentation is aerobic glycolysis in humans, but whether there is an ontological analogy of Warburg effect in cancer and (embryonic) stem cells is unknown, as to our knowledge, the potential existence of Warburg effect in embryonic stem cells remains uninvestigated. As we discuss later, though, indirect indication is that both embryonic and cancer stem cells express the isoform M2 of pyruvate kinase, an enzyme which modulates flux of carbon through the later steps of glycolysis (Vander Heiden et al., 2009).

Approaching a critical review of the former hypotheses, it can be drawn that most experimental approaches have failed to taken into consideration the micro-environment of cancer cells. Concomitantly, they have not investigated the effect of basal release of ATP from the stroma cells, a process which exercises autocrine or paracrine signaling (Corriden and Insel, 2010). An inverse scenario has been recently proposed by Pavlides et al. (2009) on the "reverse Warburg effect" in which epithelial cancer cells induce the Warburg effect in stromal neighboring fibroblasts, which secrete lactate and pyryvate that are later on received by the cancerous cells.

The aim of the present article is not to analyse separately the connection of every enzyme of the glycolytic or tricarboxylic acid cycle with carcinogenesis nor is it to explain how inhibition of these enzymes could offer protection against cancer, as the former parameters have been thoroughly analysed elsewhere (Scatena et al., 2008; Furuta et al., 2010). Rather, it is to offer an up-to-date critical analysis of the enigmatic relationship between the cancer phenomenon and the function of metabolic genes.

Tricarboxylic Acid Cycle Regulatory Genes: an Emerging Role in Cancer

Mutations in metabolic genes of the tricarboxylic acid cycle have been implicated in cancer, including succinate dehydrogenase (SDH) and fumarate hydratase mutations in paraganglioma/ pheochromocytoma and leiomyosarcomas/ renal cell cancer, respectively, in which the vast accumulation of the metabolites (succinate and fumarate) lead to inhibition of the HIF-1-suppressing enzyme proline hydroxylase (Cervera et al., 2008; Thompson, 2009). In relation to paragangliomas, in particular, mutations have also been reported for a recently identified protein, the Sdh5, which is required for SDH-dependent respiration (Hao et al., 2009). Apart, however, from HIF-activation, mutation-induced SDH inactivation might be correlated with an increased capacity for adherence to extracellular matrix proteins, implicating a potential role in the first steps of tumor metastasis (Cervera et al., 2008).

Approximately 15% of glioblastomas and more than 70% of astrocytomas and oligodendrogliomas present mutations in the amino acid 132 or amino acid 172 in the isocitrate dehydrogenase 1 (IDH-1) or 2 (IDH-2), respectively (Yan et al., 2009). In parallel, R132H

mutations in IDH have been recently implicated in acute myeloid leukemia (Mardis et al., 2009). Dang et al., (2009) have shed light on the mechanism of the monoallelic mutations of IDH. These mutations, resulting in a loss of NADPH production by isocitrate, provide the IDH enzyme with a gain-of-function ability to convert in a NADPH-dependent reduction α-ketoglutarate to 2-hydroglutarate, which is by both in vivo and clinical data implied to be oncogenic. On the other hand, since α-ketoglutarate levels are reduced in mutant IDH enzymes, proline hydroxylase activity and thus production of the degraded HIF-1-OH is not enhanced (Zhao et al., 2009). What remains as an unsolved question is why glioma patients with IDH mutations have a better prognosis (Yan et al., 2009).

Pyruvate Kinase: When it Comes to Play

Pyruvate kinase (PKM) being the enzyme converting phosphoenolpyruvate to pyruvate and thus regulating the rate-limiting final step of glycolysis has two specific isoforms; the adult isoform PKM1, promoting oxidative phosphorylation and the PKM2 isoform, which is expressed in embryonic and cancerous cells and which promotes aerobic glycolysis. The above isoforms are the result of a mutually exclusive alternative splicing of the PKM pre-mRNA, corresponding to inclusion of either exon 9 (PKM1) or exon 10 (PKM2) (Ferguson and Rathmell, 2009; David et al., 2010).

David et al., (2010) show that c-Myc contributes to upregulation of the transcription of three proteins (PTB, hnRNPA1 and hnRNPA2) which repress exon 9 leading in the inclusion of exon 10 and thus in a low PKM1/ PKM2 ratio. They further push forward this novel finding by demonstrating that in gliomas, c-Myc, PTB, hnRNPA1 and hnRNPA2 overexpression is related to PKM2 expression.

Moreover, short hairpin RNA-mediated inhibition of PKM2 and switching to PKM1 has been connected to inhibition of the Warburg effect, namely lactate production, as well as to a reduced capacity of tumor formation in mouse models. To address whether PKM2 except from being essential is also adequate for tumor formation, authors propose that PKM2 is rather a contributing factor- a provider of glucose metabolites in anabolic processes- than a "solo player" (Christofk et al., 2008a).

However, glycolysis-unrelated roles of PKM2 have emerged. The same as above laboratory found that PKM2 is the only isoform of pyruvate kinases to be a phosphotyrosine-binding protein and the latter is suggested not to be limited to the consequent release of the PK allosteric activator fructose-1,6-bisphosphonate (F1,6P) (Christofk et al., 2008b; Ferguson and Rathmell, 2008). Expanding these findings, another study pointed that PKM2 is inhibited by phosphorylation of its tyrosine residue 105 by specific isoforms of fibroblast growth factor receptor, and that it is this formation which disrupts binding of the F1, 6P. This type of phosphorylation is interestingly observed in various human cancer cell lines and a phenylalanine substitution in position 105 contributes to a reduction of cell proliferation and a reduction of the Warburg effect (Hitosugi et al., 2009). It

remains, nevertheless, elusive whether patients bearing these mutations in their cancer samples have a better prognosis or not.

Theoretically, a phosphorylation-mediated decrease in wild-type PKM2 activity should be followed by a reduction in the glucose converstion to pyruvate and lactate, but the adverse effect was observed by Hitobutsugi et al. when wild-type PKM2 was compared to the Y105F mutant. Dang (2009) proposes that putative high concentrations of pyruvate produced by the mutant Y105F PKM2 may lead to a favorisation of oxidative phosphorylation rather than lactate production and that less active phosphorylated PKM2 leads to the divertion of glucose carbons into synthesis of lipids and amino acids rather than into production of pyruvate.

Mitochondria & Metabolism: the Participant Around the Corner

In their classical paper, Hanahan and Weinberg (2000) propose that the great majority of cancers are due to six necessary alterations including evasion of apoptosis, unlimited potential for replication, angiogenesis, tissue metastasis, self-sufficiency in growth signaling, and insensitivity to growth signals. Nevertheless, but still not as a great surprise, mitochondrial metabolism namely oxidative phosphorylation seems not to be a simple bystander and this is what Warburg proposed back in the 1950s (Warburg, 1956).

All types of mtDNA alterations have been found in cancer samples and reviewed elsewhere. More striking, though, is the evidence that several mitochondrial respiratory enzyme complexes are found to be significantly lower in a spectrum of human cancers and that this decrease through correlating with aggressiveness is leading to an increase of cancer invasiveness (Lee and Wei, 2009).

In addition, not only dysfunction of mitochondrion may lead to activate nuclear transcription factors through the so-called "retrograde response" via an elevated animutation can increase the capacity of tumor cells for metastasis (Ishikawa et al., 2008; Lee and Wei, 2009).

Metabolism & Cancer: Novel Pieces of the Puzzle

Diabetes, primarily expressed by hyperglycemia, is linked to an increased risk for cancer development, particularly pancreatic, liver and colon cancer (Ogunleye et al., 2009). Increased levels of glucose contribute to increased levels of gene mutagenesis in human lymphoblastoid cells predisposing to oncogenesis, although glycation-mediated protein inactivation could play an additional role (Zhang et al., 2007).

On the other hand, GLUT-1, which encodes for glucose transporter-1, is found highly upregulated in cells bearing KRAS or the mutually exclusive BRAF mutations, and endowed these cells with a selective advantage in low glucose conditions. It was also elegantly reported that approximately 4% of wild-type cells which survived in hypoglycemic conditions had obtained novel KRAS mutations and thus it was shown for the first time that hypoglycemic conditions may lead to acquisition of mutations presenting a selective advantage (Yun et al., 2009). This selective advantage is rather critical in cancer cells, which by having increased steady states of endogenous $O_2.-$ and by thus exhibiting increased glucose metabolism to compensate for excess metabolic production of reactive oxygen species, are far more susceptible to oxidative stress and cell death induced by glucose deprivation (Aykin-Burns et al., 2009). In line with this, in Saccharomyces cerevisiae, it was reported that the aerobic glycolysis namely Warburg effect inhibits oxidative stress-mediated apoptosis, also though reduction of decreased mitochondrial respiration (Ruckenstuhl et al., 2009).

Extracellular Matrix Detachment and Glucose Metabolism

In an elegant study, Schafer et al., (2009) indirectly questioned how cancer cells can overcome anoikis and demonstrated that overexpression of ERBB2 can reverse extracellular matrix (ECM)-induced ATP reduction through stabilization of EGFR and phosphatidylinositol-3-OH kinase activation. More challenging though is that antioxidants contribute to the survival of cells with lack of attachment to ECM, an observation which underlines that antioxidants may in fact act also as tumor promoters. Parallely, stimulation of the pentose phosphate pathway leads to reduction of free radicals and thus to the promotion of cell survival (Schafer et al., 2009). In fact, glucose-6-phosphate dehydrogenase (G6PDH) levels, implicated in production of NADH levels and nucleotide synthesis, are found to be increased during cell cycle in cancer cells both in vivo and in vitro (Frederiks et al., 2008; Meadows et al., 2008; Vizán et al., 2009). Finally, p53 regulation of glycolysis could also lead to changes in the flux towards the pentose phosphate pathway (Vousden and Ryan, 2009).

Threonine Metabolism and Cancer

Recent data revealed that mouse embryonic stem (ES) cells highly express the gene of threonine dehydogenase (TDH), by which the essential amino acid threonine is converted to glycine (for purine biosynthesis) and acetyl-CoA (for tricarboxylic cycle). Notably, not only ES cells were critically dependent on threonine, but also threonine depletion reduced DNA synthesis (Wang et al., 2009). Since embryogenesis and oncogenesis might share common features including the role of stem cells (Gupta et al., 2009), an analogy of the above experiments to cancer cells would be appealing. An old study reported apparently decreased TDH hepatic activity in animals with different types of cancer (Tshudy et al., 1964). In humans, the TDH pathway accounts for only 10% percent of total threonine metabolism (Darling et al., 2000), although later reports indicate that humans seem to be incapable of producing an active TDH enzyme (Edgar, 2002). Whether this consists a selective advantage remains uninvestigated (Wang et al., 2009). Parallely, metabolomic studies on different human cancer types have not identified alternated levels

of threonine or its metabolites 2-amino-3-ketobutyrate, glycine or acetyl-CoA (Sreekumar et al., 2009).

Epithelial-mesenchymal transition and metabolism

Data from Weinberg and Brisken laboratory have shown that cells that have undergone epithelial-mesenchymal transition (EMT) obtain properties and demonstrate markers of stem cells, and form moreover tumors more efficiently (Mani et al., 2008).

Novel insights into the link of EMT with metabolism have been raised since overexpression of phosphoglucose isomerase (PGI), an enzyme for the interconversion of glucose-6-phosphate and fructose-6-phosphate, is found to induce mesenchymal-like cells and to disrupt the E-cadherin/ β-catenin cell adhesion complex. Inversely, also, siRNA-mediated downregulation of PGI leads to mesenchymal-to-epithelial transition (Funasaka et al., 2009). Inversely, down-regulation of PGI results in mesenchymal-to-epithelial transition at least in human lung fibrosarcoma (Funasaka et al., 2007). While hypoxia is proven to induce expression of HIF-1 and consequently that of PGI, the inverse relation remains indefinable. Potentially, PGI mediates per se EMT due to its manifold action as a cytokine including autocrine motility factor with mitogenic and differentiation properties (Funasaka et al., 2005). Parallely to hypoxia, high concentrations of glucoses contribute to EMT transition potentially through PGI, a phenomenon reversed by the overexpression of BMP-7 and hepatocyte growth factor (Yu et al., 2009).

On the other side, hyperoxia induces MET in vivo and leads to tumour reduction though decreases in tumor cell proliferation and reduction of tumor blood vessels and of collagen fibrils. Notably, following hyperbaric treatment glycolytic enzymes hexokinase II and glyceraldehyde-3-phosphate dehydrogenase as well as lactate dehydrogenase were reduced. Authors although failing to provide evidence on the underlying mechanism of MET, suggested that oxygen per se can induce MET (Moen et al., 2009), a note which would be implicative of redox signaling in MET and EMT. In fact, an elegant experiment by Cannito et al., (2008) revealed that hypoxia-induced early MET phenomenon was dependent on transient intracellular increased generation of ROS and very early inhibition of glycogens synthase kinase-3-beta, while late migration and invasiveness were mediated by HIF-1a.

ATP citrate lyase and cancer

ATP citrate lyase (ACL) catalyses the conversion of citrate to oxaloacetate and acetyl-CoA and ACL production of acetyl-CoA for de novo fatty acid synthesis is crucial for the proliferation of glycolytically-converted tumor cells (Wellen et al., 2009). In fact, ACL activity is found increased in cancer cell lines, in which phosphorylated ACL forms are overexpressed, wheareas inhibition of ACL leads to the arrest of cancer growth (Migita et al., 2008).

Wellen et al., (2009) opened a new window in mammalian epigenetics by demonstrating that histone acetylation is dependent on ACL and that ACL is required for increases in histone acetylation in relation to growth factor stimulation as well as during differentiation. This key metabolic enzyme can explain novel findings

suggesting that concentration of fatty acids and glucose affect the acetylation process which now is not considered restricted to histone modification but contrary, is broaden to the acetylation of almost every enzyme in glycolytic and tricarboxylic acid cycle (Zhao et al., 2010).

Sirtuins and cancer: the Janus face?

Mammals have seven members of sirtuins, SIRT1 to SIRT7, and NAD^+ is required for all sirtuins enzymatic activity, irrespective of whether they exhibit solely deacetylase activity (SIRT 1, 2, 3, 5, 7), solely ADP-ribosyltransferase activity (SIRT 4) or both (SIRT 6). The most clearly defined role of sirtuins refers to metabolic homestasis, in which SIRT1 is found to promote PGC-1a-mediated gluconeogenesis and fatty acid oxidation (Yu and Auwerx, 2009). Contrary, glycolysis consumes NAD+ during the metabolism of glyceraldehyde to 1,3 diphosphoglycerate and NADH, whose accumulation results in decrease of sirtuins activity (Kassi and Papavassiliou, 2008).

Proven the role of sirtuins in energy homeostasis and the need of new drugs for reducing diabetes, the elucidation of sirtuins function in cancer is essential in order to avoid detrimental side-effects. The connection of sirtuins with cancer and cancer metabolism illustrates its double role both in the deacetylation of histones (H1, H3, H4) and tumour-related transcription factors (p65 component of NFϰB, p53, DNA repair factor Ku70, FOXO proteins) as well as in the regulation of glucose metabolism, known to be alternated in cancer (van Leeuwen and Lain, 2009). To our knowledge, no study has investigated the role of sirtuins in tumour-specific metabolism, although it would be interesting to investigate how potential changes in NAD^+ levels in cancer could affect sirtuins expression (van Leeuwen and Lain, 2009).

A plethora of studies has searched sirtuins effects on transcription factors and histones in cancer and have been reviewed elsewhere (for review, Deng, 2009; van Leeuwen and Lain, 2009; Yu and Auwerx, 2009). Two major categories of data have contributed to characterize SIRT1 as a tumour promoter (for more profound analysis, on whether sirtuins are tumour promoter or suppressors, please see above reviews). First is the noted overexpression of SIRT1 in some cancerous samples (reviewed in Deng, 2009). A conciliatory approach may be derived from a recent study in which SIRT1 levels were found to be highly increased in osteosarcoma cells, but interestingly administration of resveratrol and isonicotinamide, both SIRT1 activators, led to inhibition of cell growth (Li et al., 2009). More interestingly, another study reported that although SIRT1 per se did not suppress tumour formation, a relevant reduction by resveratrol was not dispensable but still highly dependent on SIRT1 (Boily et al., 2009). It is notable that Canto and Auwerx (2009) have suggested that SIRT1 activation may be necessary for resveratrol action as a downstream result of AMPK activation.

Second data suggesting the tumour promoter "face" of sirtuins is the fact that SIRT1 deacetylates p53, albeit it remains unknown whether sirtuins deacetylate the lysine residues in p53 which are critical for its activity (van Leeuwen and Lain, 2009). Nevertheless, a recent study

challenged previous considerations that SIRT1 promotes oncogenesis through deacetylating p53, as p53$^{+/-}$ mices overexpressing SIRT1 were at less risk of developing irradiation-induced thymic lymphoma and also had a mean 35% increased lifespan (Oberdoerffer et al., 2008). Parallely, SIRT1$^{+/-}$; p53$^{+/-}$ mice developed tumours in multiple tissues, the majority of which preserved one wild-type allele of SIRT1, implying that an appropriate dose of SIRT1 is critical for repressing tumour formation (Wang et al., 2008). In addition, SIRT1 was recently reported to deacetylate a well-known tumor promoter, c-Myc, leading to its decreased stability (Yuan et al., 2009).

Notably, as a response to oxidative stress, SIRT1 may relocalise from silencing major satellite repeats and other genes in order to physically associate with DNA double-strand breaks, illustrating that SIRT1 overexpression can repress genomic instability (Oberdoerffer et al., 2008). Inversely, absence of SIRT1 expression was found to promote genetic instability (Wang et al., 2008).

In colon cancer cell lines, SIRT1 overexpression significantly suppresses tumour formation and proliferation by deacetylating beta-catenin and by promoting its cytoplasmic localization (Firestein et al., 2008). On the other hand, in human colon cancer samples, SIRT1 overexpression, found in approximately 40% of samples, correlated with high tumour grade and was related to individual CpG islands but not to global DNA methylation (Nosho et al., 2009). The authors noted that a difference in patient cohorts or false positive/ negative results in immunochemistry could explain why no association of SIRT1 and β-catenin expression was reported in the latter study. Additionally, Boily et al., (2009) reported low levels of SIRT1 expression in the intestine and in order to explain the repressive effects in Firestein's et al work, they suggested that SIRT1 may be a "conditional tumour suppressor", the activity of which depends on achieving a threshold of enzymatic activity. Finally, SIRT1 expression was found to be negatively correlated with the expression of deleted-in-breast-cancer-1 gene (Escande et al., 2009).

In a relatively recent study far from studying effects of SIRT1 on different substrates, Narala et al., (2008) showed that SIRT1 inhibition led to an increase in telomerase activity and to promotion of cell growth. Notably, cells lacking SIRT1 and being under glucose deprivation presented an earlier increase in total and phospho-AMPK, which could permit survival in the above ATP-limiting conditions. These are in line with findings by Canto et al., (2009) suggesting the opposite direction, that AMPK induces SIRT1 activity by increasing cellular NAD$^+$ levels.

With regards to a potential involvement of autophagy in cancer metabolism, SIRT 1 is shown to be essential for starvation-induced autophagy through forming complexes with and deacetylating different components of the autophagy machinery, namely Atg5, Atg7 and Atg8 (Lee et al., 2008). However, although molecules regulating of autophagy are also implicated in tumour suppression (Levine and Kroemer, 2008), more elegant experiments are essential in order to determine the role of SIRT1. Notably, the complexity is further strengthened by the fact that tumour cells may use autophagy to survive from metabolic stress (Jin and White, 2008), although other reports note that autophagy-related survival in some experiments may not cause tumour promotion (Levine and Kroemer, 2008).

While SIRT1 is the most well investigated sirtuins on this subject, the recent elucidation of the physiological role of other sirtuins permits preliminary assumptions on their potential role in cancer to be formed. SIRT1 and SIRT3 deacetylate and thus activate cytoplasmic and mitochondrial acetyl-CoA synthetase, respectively (Yu and Auwerx, 2009). The latter enzyme is found to play an important role in acetate-dependent lipid synthesis for cancer cells growth exhibiting a low-glycolysis phenotype and to be a highly expressed bi-directional enzyme in hypoxic tumor cells, in which its inhibition results to increased cell death (Yoshii et al., 2009; Yun et al., 2009). These results which may indicate an indirect role of SIRT3 in tumour progression are contradictory with previous findings implicating SIRT3 in tumour reduction (Allison and Milner, 2007). A potential explanation could be that SIRT3 may have more than one substrates.

SIRT4 is known to transfer ADP-ribose from NAD to glutamate dehydrogenase (GDH) and to inhibit its activity (Haigis et al., 2006; Nakagawa et al., 2009). GDH converts glutamate to a-ketoglutarate and vice versa and particularly expressed in astrocytes, where glutamate act as an important neurotransmitter. Thus, inhibition of GDH by SIRT4 could potentially exercise similar effects to those by RNAi-mediated GDH inhibition and consequent apoptosis in neuroblastoma cells (Choi et al., 2009). In addition, SIRT1 is found to promote a FOXO3a-mediated differentiation of neuroblastoma cells, implying a tumour suppressive role in this cancer form (Kim et al., 2009).

SIRT5 is found to be a mitochondrial matrix NAD-dependent deacetylase which specifically deacetylates and thus activates the carbamoyl phosphate synthetase (CPS) 1, an enzyme catalyzing the reaction of ammonia with HCO$_3$- and ATP by producing carbamoyl phosphate that is then converted to urea (Nakawaga et al., 2009). Although CPS 2 has been reported to be elevated in cancer samples a long time ago (Aoki and Weber, 1981), no data seem to exist on CPS 1. Indirect evidence from colon cancer samples suggest an increase in urea cycle metabolites (Denkert et al., 2008), yet the implication of SIRT5 in these environment remains uninvestigated.

Regarding SIRT6, it is found to bind to NF-ϰB subunit RELA, to act as a deacetylase of histone H3K9, to consequently destabilize RELA from chromatin at NF-ϰB target genes and thus to inhibit NF-ϰB signaling (Kawahara et al., 2009). Given that NF-ϰB signaling plays a pivotal role in carcinogenesis (Yu et al., 2009), it would not be paradoxical to expect a non tumour-promoting role of SIRT6. In accordance with this statement, SIRT6 has been recently found to function as a corepressor of HIF-1, known to be involved in tumor-cell glycolysis and angiogenesis (Thompson et al., 2009; Zhong et al., 2010).

SIRT7 seems to be a positive regulator of RNA polymerase expression and activity, and to promote cell survival (Ford et al., 2006). All cancer upregulated kinases would promote Pol1 transcription, however, a role of SIRT7 remains still elusive (Drygin et al., 2009).

In order to explain the "Janus phase" of sirtuins in

cancer and the underlying inconsistency between studies, several mechanisms have been proposed. Differences could be due to yet undiscovered pathways controlled by sirtuins, to different SIRT1 inhibitors tackling different signaling pathways or to different cell types suggesting tissue-specific results. The latter mechanism interestingly could be explained by differenct circadian rythms of differenct cells (Jung-Hynes and Ahmad, 2009). Additional evidence for the tissue-specific regulation come from experiments with Salermide, a SIRT1 inhibitor, which affected the growth of leukemia, lymphoma and colon cancer cells, but not that of breast cancer cells (Lara et al., 2009). Moreover, since cells at the center of the tumours live in an environment with metabolic stress formed by low pH, hypoxia and nutrient deprivation and since at this region, autophagy is localized, (Jin and White, 2008), it would be tempting to speculate that SIRT1 expression would be increased at these regions, explaining at least partially the noted differences.

Fatty Acid Metabolism: the Oboe of the "Metabolic Orchestra"?

New evidence on the epidemiologically revealed connection of obesity with cancer has come recently into light, as both genetic and dietary obesity seems to contribute to the formation and progression of hepatocellular carcinoma in vivo. These mechanisms seem to be mediated through IL6, TNF and TNF 1 R signaling (Park et al., 2010).

On a molecular level, and moving from the catabolic Warburg effects to the "enorchestration" of anabolic processes, cancerous cells demonstrated a profound increase in de novo fatty acid synthesis, in contrast to normal cells, which are considered to obtain fatty acids mainly from dietary sources (Yecies and Manning, 2010).

Nomure et al., (2010) recently showed that an enzyme responsible for hydrolyzing monoacylglycerol to release glycerol and a free fatty acid is abundantly expressed in human cancer cells where it contributes to their aggressiveness and to the increase of free fatty acids.

Concluding Remarks

The epicenter of future directions on the "cancer-metabolism arena" should be given to whether (embryonic) stem cells sharing strong similarities with cancerous cells present the Warburg effect. Another point is whether metastatic cells or circulating cancer cells, which also present the lately described tumour self-seeding phenomenon, present the Warburg effect (Kim et al., 2009).

Acknowledgments

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