Metabolism within the tumor microenvironment and its implication on cancer progression: an ongoing therapeutic target

Metabolism within the tumor microenvironment and its implication on cancer progression: an ongoing therapeutic target

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ABSTRACT

Since reprogramming energy metabolism is considered a new hallmark of cancer, tumor metabolism is again in the spotlight of cancer research. Many studies have been carried out and many possible therapies have been developed in the last years. However, tumor cells are not alone. A series of extracellular components and stromal cells, such as endothelial cells, cancer-associated fibroblasts, tumor-associated macrophages and tumor-infiltrating T cells, surround tumor cells in the so-called tumor microenvironment. Metabolic features of these cells are being studied in deep in order to find relationships between metabolism within the tumor microenvironment and tumor progression. Moreover, it cannot be forgotten that tumor growth is able to modulate host metabolism and homeostasis, so that tumor microenvironment is not the whole story. Importantly, the metabolic switch in cancer is just a consequence of the flexibility and adaptability of metabolism and should not be surprising. Treatments of cancer patients with combined therapies including anti-tumor agents with those targeting stromal cell metabolism, anti-angiogenic drugs and/or immunotherapy are being developed as promising therapeutics.

Keywords

Metabolism; tumor microenvironment; endothelial cells; immune cells; stromal cells; angiogenesis; immunosuppression

Abbreviations: Arg1, arginase 1; ASNS, asparagine synthetase; bFGF, basic fibroblast growth factor; CAFs, cancer-associated fibroblasts; CPT1, carnitine palmitoyltransferase 1; ECs, endothelial cells; ECM, extracellular matrix; FAO, fatty acid oxidation; FAS, fatty acid synthase; G6PDH, glucose-6-phosphate dehydrogenase; GLS, glutaminase; GS, glutamine synthetase; HBP, hexosamine biosynthesis pathway; HIF-1α, hypoxia inducible factor 1α; HK, hexokinase; IDO, indoleamine-2,3-dioxygenase; iNOS, inducible nitric oxide synthase; LDH, lactate dehydrogenase; MMP, matrix metalloproteinase; mTOR, mammalian target of rapamycin; NO, nitric oxide; ODC, ornithine decarboxylase; OXPHOS, oxidative phosphorylation; PCK1, phosphoenolpyruvate carboxykinase 1; PD-1, programmed death 1 receptor; PDH, pyruvate dehydrogenase; PDK1, pyruvate dehydrogenase kinase 1; PFK1, 6-phosphofructokinase; PFKFB3, phosphofructokinase-2/fructose-2,6-bisphosphatase 3; PHD: prolyl hydroxylase; PK, pyruvate kinase; PPP, pentose phosphate pathway; ROS, reactive oxygen species; TAMs, tumor-associated macrophages; TCA, tricarboxylic acid cycle; TDO, tryptophan-2,3-dioxygenase; TILs, tumor-infiltrating lymphocytes; TME, tumor microenvironment; uPA, urokinase-type plasminogen activator; VEGF, vascular endothelial growth factor.

1. INTRODUCTION

Otto Warburg started studying tumor metabolism in the first years of the 20th century and 30 years later it was proposed what we now call the "Warburg effect".[1,2] During the next years, cancer metabolism was an emerging issue in biological research, although there was a fall of interest for some years because of the boom of Molecular Biology, thought to be able to give meaningful answers to almost all questions.[3] However, many studies have been performed in the last decade due to a renewed interest in tumor metabolism, so that nowadays reprogramming energy metabolism has been considered a new hallmark of cancer.[4] By means of both classical and modern techniques, many new relevant features of metabolism of cancer cells have been discovered. Moreover, tumor cells are not alone, since a complete set of stromal and immune cells meet in the so called "tumor microenvironment" (TME), along with extracellular matrix (ECM), which provides more than an inert playground for this game.[5] These cells include endothelial cells (ECs) (vascular or lymphatic) and associated pericytes, cancer-associated fibroblasts (CAFs), and immune cells, such as tumor-infiltrating lymphocytes (TILs) (T cells, B cells and NK cells), tumor-associated macrophages (TAMs) and mast cells.[6] Studies have been usually focused on tumor and ECs metabolism. However, in the last years the metabolism of immune cells, mainly macrophages and T cells, has attracted the interest of scientific community, along with that of CAFs, due to their contribution to tumor growth. However, little is known about mast cells metabolism, and that of pericytes still remains a mystery.

Increasing knowledge about metabolism of cells of the TME will allow for the design of new therapies for cancer patients. Many compounds have already been tested for the inhibition of tumor cell metabolism, either aerobic glycolysis, glutaminolysis or other metabolic targets.[7-13] New approaches for therapy are also being developed using metabolism of stromal and immune cells as a target.[14-17]

There are many published works about metabolism of stromal and immune cells in the TME and their relationship with tumor progression. A recent review collected the effects of tumor metabolism in the TME.[18] Nevertheless, to our knowledge the relation between metabolism of the different cells of the microenvironment and tumor progression has not been well documented in a single review so far (see Supplementary Table 1). This review will try to shed light on the remarkable metabolic features of different cells of the TME and their relation with tumor progression, as well as proposing feasible therapies based on possible metabolic targets that would help in the inhibition of tumor growth and metastasis.

2. TUMOR CELLS METABOLISM: BEYOND WARBURG EFFECT

The experiments carried out by Otto Warburg in the mid-twenties of the 20th century were just the beginning of an advanced knowledge in cancer metabolism. As early as

1925, he observed a huge amount of lactic acid in rat carcinoma even when oxygen was available, a process known as aerobic glycolysis.[1] This contradicted the well-established Pasteur effect, based on the inhibition of glycolysis in the presence of oxygen.[19] Warburg also observed that malignant tumors produced more lactic acid than benign tumors.[1] Nowadays we know that the glycolytic rate can be a sign of tumor aggressiveness. For example, the non-invasive MCF7 breast cancer cell line has a lower rate of aerobic glycolysis than the highly invasive MDA-MB-231 breast cancer cell line, corresponding to lower levels of lactate dehydrogenase-A (LDH-A) and to the oxidative source of the great majority of the ATP produced by MCF-7 cells.[3,20,21] However, aerobic glycolysis is not just a sign of tumor aggressiveness, since some proliferating non-transformed cells show this metabolic characteristic too.[22] 30 years after initial Warburg's seminal observations, when many metabolic routes had been already discovered, he noticed that cancer cells could obtain similar amounts of energy by aerobic glycolysis and by oxidative phosphorylation (OXPHOS), in spite of the lower efficiency in ATP yielded per molecule of glucose provided by glycolysis.[2,23] At that moment, it was difficult to find an explanation for this fact, since high rates of tumor cell proliferation would require the production of great amounts of energy in the form of ATP molecules, and OXPHOS was the obvious road to fulfill this purpose. Now we know that, due to that high proliferation, cancer cells have a large demand of the precursors for the new daughter cells generated by mitosis, in form of nucleotides, amino acids and lipids. Thus, glucose would be diverted to the formation of acetyl-CoA for fatty acid synthesis, glycolytic intermediates for non-essential amino acids, and ribose for nucleotides.[24] This explains why many types of cancer cells switch their glucose metabolism towards aerobic glycolysis. Extracellular flux analyzers are currently very popular tools for measuring basic metabolism, since they are able to estimate OXPHOS through oxygen consumption rate (OCR) and aerobic glycolysis through extracellular acidification rate (ECAR). Nevertheless, studies with isolated tumors from mice showed that although progressive tumors have higher ECAR levels than regressive ones, their proliferation rates are similar, demonstrating that proliferation is not the only reason for aerobic glycolysis in tumor cells.[25]

The increased glucose consumption by many cancers is the basis for the use of the glucose analogue 2-[^{18}F]-fluoro-2-deoxy-D-glucose for tumor diagnostic and treatment follow-up by using positron emission tomography (PET).[26] In high contrast with the affirmation that all tumor cells rely mostly on aerobic glycolysis, there is ample evidence that not all cancer cells obey this rule. For example, glutamine is the major energy source for cervix adenocarcinoma HeLa cells, and Gentric et al. have reported some examples of oxidative tumors.[27,28] Furthermore, oxidative and glycolytic cancer cells can co-exist within the same tumor, and a lactate shuttle is established between both of them. [29] Lactate uptaken by oxidative cancer cells (either from other cells or from the circulation) can provide carbon skeletons to be incorporated to the tricarboxylic acid cycle (TCA) in order to obtain energy.[30] We would like to emphasize that in the next sections and figures of this review we will not make a distinction between oxidative and glycolytic tumor cells for the sake of simplicity. It should be

taken into account that different metabolic events here represented in the same tumor cell might be occurring in different cancer cells, though.

Nonetheless, other substrates different to glucose are also differentially consumed by tumors as well. In particular, glutamine, the most abundant circulating amino acid in blood, has a major role regarding tumor growth, as glucose can only provide carbon skeletons for scaffolds of new molecules and glutamine would serve as a nitrogen source.[31] In fact, glutamine is a non-essential amino acid for non-transformed human cells but it turns into an essential amino acid for tumor cells.[12] Moreover, a host to tumor net flux of glutamine has been confirmed in mice inoculated with Ehrlich ascites tumor cells, enabled by an increased contribution made by the host tissues to circulating glutamine during tumor development.[32,33] We will discuss this issue in a later section of this review.

Almost 30 years ago, our group found out that Ehrlich ascites tumor cells, grown under steady state conditions, utilize both glucose and glutamine, producing two moles of lactate per mole of glucose, and one of glutamate and ammonia per mole of glutamine consumed.[34] That means that cancer cells are able to use glucose and glutamine in a completely dissipative way. Both nutrients are important, as they lead to ATP production and provide intermediates for macromolecular synthesis. The roles of glutamine in intermediary metabolism have already been revised.[35] Additionally, glutamine can be used for synthetizing the non-essential amino acids alanine, serine, arginine and proline and also fatty acids, although glucose is the major lipogenic substrate, as seen in glioblastoma cells.[36,37] It is important to remember that glutamine can lead to lactate production through glutaminolysis. So, aerobic glycolysis is not the only way a tumor cell possesses to produce lactate, whose excretion out of the cell was first thought to be a mechanism to eliminate the pyruvate excess.[23] However, lactate would have many roles in benefit of tumor progression that will be discussed in other sections of the present review. Likewise, ammonia was also thought to be just a toxic waste product. Nevertheless, it has been recently shown that this metabolite can be recycled to generate amino acids through glutamate dehydrogenase (GDH) activity, providing a nitrogen source to the tumor.[38]

Metabolic profiling depends on cell distribution, as cancer cells within the oxygenated periphery may consume and oxidize the lactate resulting from aerobic glycolysis by cells in the hypoxic area.[39] Besides, cancer metabolic phenotypes are usually defined by the origin of the tissue, epigenetic drivers, aberrant signaling, and the TME.[40] Indeed, genetics, epigenetics and metabolism interact with one another and, as a result, tumor heterogeneity is the overall result of all these changes at different levels.[41] A previous review of tumor metabolism contributed by our group focused its attention in the genetic regulation of tumor metabolism. The key roles played by c-myc, K-Ras and p53 are well documented. For example, c-myc oncogene promotes expression of LDH-A, the glutamine transporter SLC1A5 and GLS glutaminase (associated to tumor malignancy), and K-Ras stimulates glucose uptake, lactate production and canalization

of glutamine carbons to the Krebs cycle, whereas tumor suppressor gene p53 induces GLS2 glutaminase expression (typical of non-proliferative cells), OXPHOS and fatty acid oxidation (FAO), and diminishes expression of glucose transporters and some of the key glycolytic enzymes.[42]

Epigenetics plays also a role in tumor metabolism. For example, 2-hydroxyglutarate (2-HG), a product of the reaction catalyzed by a mutated isocitrate dehydrogenase 1 (IDH1), inhibits the binding of α-ketoglutarate (α-KG) to tet methylcytosine dioxygenase 2 (TET2) and lysine demethylase 3A (KDM3A), two epigenetic enzymes, impairing their function.[43] Another example is nitric oxide (NO), also able to drive epigenetic modifications related with tumorigenesis.[44]

Less attention has been paid to studying the role of fatty acids in tumor growth, since glucose and glutamine are considered the major sources of energy in these cells. A relationship between glycolysis and FAO has been found in tumors, since highly glycolytic cell lines present a low lipid oxidation and *vice versa*.[45,46] Some tumors lack carnitine palmitoyltransferase 1a (CPT1a) activity, a rate-limiting enzyme of FAO.[47] In various tumor cell lines, rates of oxidation of glucose higher than those of palmitate have been documented.[48] However, it has been shown that highly proliferative cancer cells have a strong lipid avidity, increasing the uptake of exogenous lipids or promoting lipogenesis and cholesterol synthesis.[49] Fatty acid synthase (FAS) is overexpressed in several types of cancer.[50–52] Transcription factors SREBP1 and SREBP2, involved in fatty acid and cholesterol biosynthesis, are also overexpressed in many tumors.[53] On the other hand, prostate tumors display a low rate of glucose utilization; they rather have a high rate of fatty acids uptake and overexpress some β-oxidation enzymes.[53] It has been shown that leukemia cells require this metabolic route for proliferation and survival.[54] Additionally, there is some controversy about the role of fatty acids on metastasis and invasiveness. A published study found an inverse relationship between expression of CD36, a known transporter of long fatty acids, and the metastatic potential of tumors, whereas the authors of a more recent paper postulate a positive role of CD36 in metastasis.[55,56]

Other metabolites could also play essential roles in tumor metabolism. The role of asparagine in cell survival has been well-known for many years, and several studies are being carried out nowadays regarding the importance of this amino acid. The presence of asparagine is essential for maintaining cell viability in glutamine-depletion conditions, and inhibition of asparagine synthetase (ASNS), an enzyme that catalyzes the conversion of aspartate and glutamine into asparagine, leads to cell death even in a glutamine-rich media.[57] Therefore, depleting asparagine and inhibiting ASNS expression seems to be a way to stop tumor growth. Treatment with the enzyme asparaginase, which is able to undermine asparagine levels in the media, has been carried out in leukemia and lymphomas since the discovery of its anti-cancer effect in 1963.[58] Later, it would be known that asparaginase treatment was effective due to the null or low expression of ASNS in these tumors.[59,60] Nevertheless, most solid tumors

present ASNS expression and therefore depletion of glutamine is also important for asparaginase-dependent therapy in ASNS-expressing tumors.[61,62] Indeed, a study determined that glioblastoma cells that are not sensitive to glutamine deprivation are also insensitive to asparaginase treatment, but the treatment affected glioblastoma cells sensitive to deprivation of this amino acid.[63] This may be due to the fact that most asparaginases also present glutaminase activity.

There are other amino acids that are essential for tumor growth and progression as well. Serine can be synthetized from glycolytic intermediates and later converted into glycine. Both amino acids are necessary for protein, nucleic acid and lipid synthesis. Serine can contribute to the formation of other metabolites by anaplerosis, being necessary for proliferation. Glycine, which may also derive from threonine, is related to folate metabolism (essential for tumor progression), to DNA methylation, and to the redox balance maintenance.[64,65] Indeed, expression of PHGDH (phosphoglycerate dehydrogenase), the first enzyme in serine synthesis, is normally upregulated in triple-negative breast cancer, evidencing the importance of this amino acid for these tumors.[66] In contrast, metabolism of other amino acids can be toxic for tumor cells. For example, proline oxidase (PRODH), the first enzyme in the catabolism of proline, is induced by p53.[67] Expression of PRODH leads to cell cycle arrest and apoptosis in tumors, and it has been seen that c-myc inhibits its function.[68]

In addition to all this, other metabolites are also important for tumors. NO is the product of the enzymatic reaction catalyzed by nitric oxide synthase (NOS), which uses arginine as substrate, as well as NADPH. Thus, the pentose phosphate pathway (PPP) would provide the reducing agent necessary for synthetizing NO. In hypoxic tumors, hypoxia inducible factor 1α (HIF-1α) interacts with IFN-γ thus inducing the expression of inducible NOS (iNOS).[69] NO produced and secreted by tumor cells reprograms stromal cells to support tumor progression, although high concentrations has been shown to induce apoptosis, and it also helps drug resistance and migration of cancer cells.[70-72] Moreover, NO modulates metabolism of tumor cells, inhibiting prolyl hydroxylase 2 (PHD2) and OXPHOS, hence promoting a glycolytic metabolism.[73,74] Furthermore, S-nitrosylation is a mechanism of posttranslational protein modification mediated by NO and implied in modulating the activity of several oncogenic signaling cascades and metabolic enzymes.[69]

Last but not least, polyamine synthesis has been known to be essential for tumor progression since the late sixties.[75] High levels of intracellular polyamines have been shown to increase cell proliferation, decrease apoptosis, enhance expression of genes affecting tumor invasion and metastasis, and they are also related to angiogenesis.[76] The synthesis of these macromolecules requires conversion of arginine to ornithine through arginase activity. Then, ornithine is decarboxylated to produce putrescine, the first polyamine, in a reaction catalyzed by ornithine decarboxylase (ODC), and spermidine and spermine are synthetized using decarboxylated S-adenosylmethionine (dcSAM) as an aminopropyl group donor.[77] ODC was described as a proto-oncogene as soon as

1992, and ODC levels are higher in tumors than in non-proliferating tissues.[78,79] Moreover, several oncogenes, such as myc and K-Ras, are responsible for augmented polyamine synthesis and decreased polyamine catabolism, thus promoting tumor progression.[80–82] Interestingly, NO is able to inhibit ODC by nitrosylation.[83] Polyamine synthesis in tumors has been classically suppressed by treatment with difluoromethylornithine (DFMO), an inhibitor of ODC.[84] Recent research has found that mammalian target of rapamycin complex 1 (mTORC1) sustains polyamine synthesis in tumors through overexpression of S-adenosylmethionine decarboxylase 1 (AMD1), the enzyme responsible for SAM decarboxylation.[85]

The different metabolic features of tumor cells mentioned here are collected in Figure 1. Taking into account all this information, it cannot be said that all tumor cells rely just on aerobic glycolysis for its growth and progression. In fact, this depends more on the kind and stage of the tumor, as well as on its microenvironment. Metabolism of different cells of this TME will be presented throughout this review, along with a recapitulation of the feasible reasons and/or consequences of those metabolic features in cancer disease.

3. METABOLISM OF CELLS AT THE TUMOR MICROENVIRONMENT

3.1. Endothelial cells

ECs are the most studied stromal cells in the TME, since they are responsible for the angiogenic process. Angiogenesis is the formation of new blood vessels from the pre-existing vascular bed. Pathological activation of angiogenesis in tumors (a process called tumor angiogenesis) allows them to grow and metastatize. This angiogenic switch is controlled by pro- and anti-angiogenic molecules secreted from different cells of the TME.[86] As we discuss throughout this review, metabolic pathways regulate some of these angiogenic molecules, representing promising targets to modulate tumor angiogenesis. Therefore, targeting metabolism to inhibit tumor proliferation could be also a way to modulate the angiogenic process.

Regarding EC metabolism, there are some discrepancies among published data. Back in 1991, Spolarics et al. determined that rat liver ECs rely predominantly on aerobic metabolism rather than glycolysis, with 45% of total ATP produced by oxidation of palmitate, and 26% derived from glutamine.[87] Three years before, Leighton and colleagues measured glutaminase activity in bovine pulmonary ECs, and found that it was almost 20-fold higher in comparison with that of rat lymphocytes, giving a major importance to glutamine metabolism in these cells. They also recognized some relevance to FAO, since CPT1a showed an elevated expression. However, in contrast with the results from Spolarics's group, their data showed high activity of some key glycolytic enzymes, such as hexokinase (HK), 6-phosphofructokinase (PFK1) and

pyruvate kinase (PK), suggesting that glycolysis could play an important role in EC metabolism.[88] Indeed, other groups found glycolysis to be predominant in bovine cavernous, rat coronary and human umbilical vascular ECs (HUVEC) even in the presence of oxygen.[89-91] From these and other data, it has been proposed that ECs rely on glutamine and fatty acid metabolism when the supply of glucose decreases.[92] Most of these differences observed in bibliography could be probably due to different isolation and culture conditions of ECs, affecting their proliferation rate and their metabolism.

The interest on EC metabolism was pushed into background for some years, until 2013, when Peter Carmeliet's laboratory found interesting data regarding the role of phosphofructokinase-2/fructose-2,6-bisphosphatase 3 (PFKFB3) activity in EC metabolism and angiogenesis. In their experiments, they observed that ECs isolated from several tissues were highly glycolytic, >200 fold-higher compared to oxidation of glucose, glutamine or fatty acids in the same cells, generating up to 85% of the total cellular ATP content only through this pathway.[93] In addition, a reported low OCR in HUVEC may indicate that they rely more on glycolysis than on OXPHOS.[94] These observations agree with previous results from other groups and disagree with other available data, as seen above.[87-91]

PPP is also important for ECs, since it leads to the formation of reduction equivalents as NADPH, induces the synthesis of NO, a pro-angiogenic factor, and prevents the formation of reactive oxygen species (ROS). Indeed, studies in ECs have shown that an overexpression of the limiting enzyme of the PPP, glucose-6-phosphate dehydrogenase (G6PDH), results in an increase of NO synthesis, whereas its downregulation drives to an elevation in ROS levels.[95] On the other hand, a part of the glucose metabolic flux is derived to the hexosamine biosynthesis pathway (HBP), essential for the N-linked glycosylation process. HBP may play a role in angiogenesis switch on, since VEGFR2, the key vascular endothelial growth factor (VEGF) receptor involved in tumor angiogenesis, has to be N-glycosylated to become fully functional.[96] Regarding tumor progression, glycolysis-derived lactate has also an important role on the angiogenesis process (see section 4.2 below).

In spite of the rediscovered importance on endothelial glycolysis, glutamine metabolism is still considered to have an essential role in EC survival, as well as in angiogenesis.[97-99] However, glutamine helps EC proliferation but not migration.[100] A part of the importance of glutamine metabolism in EC survival and angiogenesis could be due to the role of this amino acid in the synthesis of polyamines, considered to be essential to EC proliferation and angiogenesis, as well as for cell survival.[101,102] In fact, in some EC lines about a 26% of ornithine, the precursor for polyamine synthesis, is formed from glutamine.[103,104] In addition, glutamine is also essential for asparagine synthesis through ASNS activity, as seen above. A recent study showed that asparagine can be uptaken from the media or synthetized by ASNS in ECs, and this amino acid has

an important role in protein synthesis, mTOR activation and endoplasmic reticulum (ER) stress suppression due to glutamine deprivation.[99]

As mentioned above, Spolarics et al. suggested that fatty acids could be important fuels for ECs, in high contrast with previous observations from other group.[87,105] A recent study from the same group that underestimated the use of fatty acids in ECs, showed later that FAO is essential for angiogenesis by promoting the *de novo* synthesis of nucleotides, thus allowing ECs to proliferate.[93,106] In fact, inhibition of CPT1a impaired angiogenesis in HUVEC.[106] One of the long chain fatty acids transporters in ECs is CD36. Inhibition of CD36 has been shown to reduce angiogenesis, but it is not clear whether this effect is due to fatty acid uptake inhibition or not.[107]

Metabolism of ECs is summarized in Fig. 1. For additional information, we encourage our readers to visit some recent reviews on EC metabolism summarizing what is known about glucose, glutamine and fatty acid fate in these cells.[107–110]

3.2. Cancer-associated fibroblasts

CAFs are the most abundant cells within tumor stroma. They are recruited by tumor cell-secreted platelet-derived growth factor (PDGF).[111] It is well known that CAFs promote tumor growth and invasion, although recently published works showed contradictory results regarding intestinal tumorigenesis.[112–114]

Although from now on we will assume the classical view, it should be clear that metabolism and signaling pathways are complex and probably there is not an absolute truth. Bearing this in mind, it has been shown that CAFs resemble myofibroblasts, as they express smooth muscle cell markers and produce transforming growth factor β (TGF-β) and stromal cell-derived factor 1 (SDF1). Additionally, CAFs express the migration stimulating factor (MSF), whose overexpression leads to Akt pathway activation, which in turn induces the mTOR signaling pathway.[115] CAFs expressing MSF showed elevated lactate secretion.[115] Since mTOR is known to enhance glycolysis, it could be proposed that MSF increases the glycolysis rate in CAFs through mTOR signaling. This high lactate secretion by CAFs is supported by the upregulation of MCT4, a lactate exporter, observed in these cells.[116] Zhang and colleagues demonstrated that IDH3α, a TCA enzyme, is downregulated in CAFs, and this situation leads to HIF-1α stabilization, resulting in a switch from OXPHOS to glycolysis.[117] As we will see below, tumor cells could as well induce this glycolysis activation. Moreover, CAFs are also able to take up lactate (secreted by tumor cells) through MCT1, a lactate importer, and to oxidize it.[118,119]

It has been shown that CAFs have a metabolic activity higher than that of other fibroblasts, since they present higher expression levels of glutamine synthetase (GS), of several glycolysis, TCA cycle and ETC gene products, and aspartate and asparagine

(both required for glutamine synthesis in these cells) transporters.[120] A summary of CAFs metabolism is presented in Fig. 1. The importance of glutamine and fatty acid synthesis by CAFs in the TME will be discussed later.

3.3. Tumor-associated macrophages

Macrophages are a population of immune cells originated from bone marrow-derived monocytes (BMDM) and exhibiting a great heterogeneity in phenotype and functions. These cells help tumors to grow and invade other tissues, promoting tumor progression also by stimulating angiogenesis and inhibiting the immune response. As in the case of CAFs, the energetic metabolism of non-tumoral macrophages has been more studied than that of TAMs.

According to the activation pathway, there are two main subtypes of macrophages: M1 macrophages, activated by the canonical pathway in response to IFN-γ and LPS stimulation, and M2 macrophages, activated by an alternative pathway in response to interleukins IL-4, IL-10 and IL-13. M1 macrophages secrete pro-inflammatory cytokines and have an anti-tumoral activity, while M2 macrophages have anti-inflammatory properties. Some authors maintain that TAMs share many, but not all, features of M2 phenotype, whereas others did not find M2 markers in TAMs.[122-125] However, IL-4 is sufficient for TAM polarization after monocyte recruitment by cytokines such as CCL2 and CSF-1.[121] Moreover, a transcriptome study determined that TAMs shared genes with both M1 and M2 macrophages.[126]

It is well-established that M1 macrophages rely largely on aerobic glycolysis, maybe regulated by itaconate.[127] M2 macrophages have not remarkable glucose consumption rates. In contrast, high FAO and OXPHOS have been found in these cells. On the other hand, M1 macrophages were found to have enhanced expression of PFKFB3 isoenzyme, whereas alternatively-activated macrophages express it at low rates.[128] Since PFKFB3 is a signal of high glycolytic rates, as happened in ECs, it can be said that M2 macrophage energy metabolism does not rely on this route.[93] Another finding suggests that succinate could be a possible indirect modulator of glycolysis. Succinate is able to inhibit PHD, leading to an increased HIF-1α stabilization, as seen before in other types of cells.[129,130] This high stabilization of HIF-1α might have two major consequences at the transcriptional level: i) HIF-1α can be translocated into the nucleus, together with the glycolytic enzyme PKM2. In the nucleus, HIF-1α forms a complex with HIF-1β and other regulatory proteins, thus acting as a transcription factor able to activate the expression of key glycolytic enzymes, such as glucose transporter GLUT-1, pyruvate dehydrogenase kinase-1 (PDK1) and LDH-A.[131] ii) The same transcription factor complex can bind to the pro-inflammatory cytokine IL-1β promoter gene and activate its transcription too.[132] In summary, succinate would have a role enhancing aerobic glycolysis and the Warburg effect, and promoting IL-1β production. Both characteristics are typical features of classically-activated macrophages. Succinate may

proceed from the anaplerotic use of glutamine, or be accumulated due to a truncated TCA cycle. Since M2 macrophages obtain energy mainly by means of FAO and OXPHOS, they do not increase succinate levels and the glycolytic pathway is not enhanced in these cells. HIF-1α can also be activated through the mTOR signaling pathway. Cytokines IL-4 and IL-13, responsible for the alternative activation of macrophages, inhibit mTOR via activation of the negative regulators TSC1 and TSC2.[133] Therefore, M2 macrophages are predisposed to oxidative metabolism through a glycolysis inhibition via mTOR/HIF-1α inactivation.

Since M1 macrophages have an anti-tumoral activity, it should be expected that TAMs have a metabolic profile more similar to that of M2 macrophages.[134] However, recent evidence reveals a high glycolytic rate in TAMs.[135,136] Moreover, an elevated eicosanoid production has been found in these cells and, on the other hand, inhibition of β-oxidation did not affect cytokine production in thyroid cancer-induced macrophages, showing the importance of FA synthesis rather than catabolism in TAMs.[135,137] Regarding amino acid metabolism, TAMs from glioblastoma or exposed to glioblastoma cells present an enhanced expression of genes related to glutamate transport and metabolism (Fig. 1).[138]

Serine has been shown to be an allosteric activator of PKM2.[28] Therefore, it could seem unlikely that M2 macrophages depend on serine utilization because their metabolism does not rely on an enhanced aerobic glycolysis. However, serine metabolism has been reported as an enriched pathway in M2 macrophages by using LC/MS-based metabolomics.[139] These last authors also found that Akt/mTORC1 pathway plays a role in increasing glucose metabolism in M2 macrophages as seen by both elevated OCR and ECAR.[139] Therefore, there are some contradictory results from different groups. However, to our knowledge there is not available data about serine metabolism in TAMs. It would be interesting to further investigate the metabolic phenotype of these cells as well as the signaling pathways that govern them.

3.4. Tumor-infiltrating lymphocytes

T cells represent the most abundant lymphocyte population involved in the adaptive immune system. There are two major types of T cells: $CD4^+$ and $CD8^+$, which are classified into different subtypes. $CD8^+$ T cells often differentiate into cytotoxic T cells (CTLs), characterized by inducing apoptosis in targeted cells. $CD4^+$ naïve T cells can become regulatory or suppressor T cells (Treg cells), which have immunosuppressive functions, or helper T cells (Th cells), a type of effector T cells that participate in the immune response. There are many subtypes of Th cells, including pro-inflammatory (Th1 and Th17 cells) and anti-inflammatory (Th2 cells) lymphocytes, according to the cytokines secreted by them. Therefore, effector T cells include CTLs and Th cells. Most of tumor-infiltrating lymphocytes (TILs) are Treg cells.

There is clear evidence that activation of T cells requires a metabolic switch similar to that undergone by many tumor cells, thus exhibiting the Warburg effect and an elevated aerobic glycolysis.[141] Something similar happens in the innate immune system.[142] Nevertheless, this metabolic switch in T cells and tumor cells is based on different causes: for T cells, this is a physiological adaptation process, whereas for tumor cells it depends on a series of intrinsic genetic mutations and external responses to the TME.[143] On the other hand, Treg and memory $CD8^+$ T cells rely on FAO and OXPHOS for its survival and differentiation. Additionally, it has been reported that *de novo* lipogenesis is required for Treg differentiation from Th17 lymphocytes (Fig. 1).[144] Effector T cells, nonetheless, can survive utilizing OXPHOS in case of glucose depletion, although cytokine production is diminished under these conditions.[145]

Phosphoenolpyruvate (PEP) has been related to the T cell receptor (TCR) activation through Ca^{2+} flux. Ho et al. observed that overexpression of phosphoenolpyruvate carboxykinase 1 (PCK1), the enzyme that catalyzes the conversion of oxaloacetate into PEP, restored PEP levels and Ca^{2+} flux in glucose-deprived T cells. This can be explained by the fact that PEP undermines the activity of SERCA, an ER calcium ATPase. Under these conditions, Ca^{2+} escapes from ER to cytosol, increasing TCR-induced Ca^{2+} flux and effector function. Moreover, TCR is able to activate glucose metabolism enhancing PKM2 activity, which in turn could contribute to PEP accumulation.[146] Thus, T cell effector function would be partially controlled by PCK1 activity.

As for other cell types, mTOR plays a crucial role in T cell metabolism. Inhibition of mTOR results in an induction of AMPK phosphorylation and, consequently, an increase of FAO, leading to differentiation of $CD4^+$ T cells to Treg. Thus, mTOR would guide these cells to Th1, Th2 and Th17 phenotypes.[147,148] Programmed death 1 receptor (PD-1), an inhibitory checkpoint receptor present in TILs, has an important role in regulating glycolysis through mTOR signaling pathways. This issue will be clarified in sections below.

Dang et al. demonstrated that HIF-1α is able to induce Th17 differentiation through transcriptional activation of RORγt. HIF-1α also binds to Foxp3, targeting it for its degradation and impairing this molecule to promote Treg development.[149] Therefore, HIF-1α would promote a glycolytic cell phenotype (by activating Th17 cells) while inhibiting oxidative metabolism (via Treg impairment).

However, glycolysis is not the only pathway necessary for T cell activation. c-Myc-dependent glutaminolysis is also essential for proper T cell effector function, as it leads to nucleotide and polyamine synthesis, necessary for supporting cell proliferation.[150] In addition, glutamine regulates T cell proliferation as well as it increases IL-2 production and IL-2 receptor expression.[151] Arginine has also been shown to improve survival and anti-tumor activity of T cells.[152] An overview of TILs metabolism is presented in Fig. 1.

3.5 The tumor microenvironment forgotten cells

There are many different kinds of cells in the TME, and the ones presented here up to now are just the more abundant and studied. Tumor-associated mast cells (TAMCs) and tumor-associated pericytes are also predominant cells in the TME and have an important role in tumor progression. However, their metabolism, as far as we know, have not been described to date.

3.5.1. Tumor-associated mast cells

TAMCs are recruited to tumors in response to stem cell factor (SCF) from tumor cells and other mast cells, as well as to VEGF from tumor cells and immune cells.[153] TAMCs secrete immunosuppressive cytokines such as TGF-β and IL-10, but their more important role in tumor progression is promoting and helping the angiogenic process.[154] TAMCs produce pro-angiogenic factors such as basic fibroblast growth factor (bFGF) and VEGF, ECM modulators such as matrix metalloproteinases (MMPs) and urokinase-type plasminogen activator (uPA), as well as chimase, tryptase and histamine.[155] Treatment with compound 48/80, which triggers histamine release, causes an angiogenic response in rats and mice.[156] Despite the importance of TAMCs in tumor progression, their metabolism has not been studied so far. Nevertheless, several studies have been carried out in non-tumoral mast cells.

In 1965, Chakravarty suggested that rat mast cells had higher glycolytic rates than oxidative ones, and some years later he and others pointed out the importance of glucose metabolism and lactate production for histamine release.[157–160] However, respiration inhibitors block histamine release, and hence energy is necessary for activation and secretion of histamine.[161] On the other hand, two different studies demonstrated the inverse correlation between glutamine metabolism and mast cell function, and tryptophan conversion to kynurenine triggers mast cell degranulation.[162-164] Kynurenine, in turn, promotes tumor invasion, further demonstrating the association between mast cell function and tumor progression.[165]

More recent works tried to shed some light on the importance of glucose metabolism for mast cell function. Sekar and co-workers studied NO metabolism in mast cells. They demonstrated that NO induced tyrosine nitration of aldolase A, inhibiting this glycolytic enzyme, with the consequent accumulation of fructose 1,6-biphosphate (FBP). This accumulation inhibited the degranulation of mast cells.[166] Enolase, the ninth enzyme of the glycolytic pathway, has been related with mast cell differentiation, and Chakravarty saw in his studies that treatment with fluoride, an enolase inhibitor, diminished the glucose-supported histamine release.[158,167] Moreover, inhibition of pyruvate dehydrogenase (PDH), the clue enzyme for the TCA, inhibits mast cell degranulation and cytokine secretion.[168] These last pieces of evidence indicate a glucose-depending mast cell function. However, other works contradict these results. FcεRI is a receptor which leads to mast cell degranulation after its ligation with IgE. FcεRI has been shown to inhibit PKM2, a process necessary for mast cell degranulation.[169] Accumulation of

FBP due to the inhibition of this enzyme ceases degranulation of mast cells. Nevertheless, these last authors mention that accumulation of FBP leads to re-activation of PKM2 and reestablishment of glycolytic normal levels, thus inhibiting mast cell function.[169] Furthermore, polyamines have been detected in mast cell granules, and treatment with DFMO diminishes histamine intracellular storage and increases PKM2 expression.[170] This fact establishes a positive relation between polyamine metabolism and degranulation of mast cells with some implication of the glycolytic pathway. Further studies should be performed in order to clarify the exact role of glucose metabolism in mast cell function and its connection with tumor progression.

3.5.2. Tumor-associated pericytes

Pericytes are responsible for morphological and functional abnormalities of tumor blood vessels, and interaction between tumor cells and pericytes has been shown to improve malignancy of glioblastoma.[171,172] Tumor-associated pericytes present greater migration and proliferation rates than normal ones, and hence they are loosely attached to ECs.[173]

Several studies have been carried out in retinal pericytes in the context of diabetic retinopathy, but without exploring glucose metabolism in pericytes.[174,175] The only work about pericyte metabolism performed to our knowledge demonstrated that lung pericytes from pulmonary arterial hypertension patients presented higher expression of PDK-1, an inhibitor of PDH, than healthy pericytes.[176] Therefore, it could be considered that normal pericytes display higher rates of OXPHOS than those of glycolysis. Nevertheless, metabolism of tumor-associated pericytes and its relation with tumor progression are yet to be studied.

4. IMPLICATIONS OF TUMOR AND ACCOMPANYING CELLS METABOLISM FOR TUMOR GROWTH AND PROGRESSION

In the previous sections, we have reviewed the main metabolic features of different cells within the TME. However, the complex interplays among these different cells and their metabolic features should be also taken into account. It is well-known that tumor stroma contributes to tumor progression.[177] Several aspects of tumor progression, such as immunosuppression and angiogenesis, depend on the metabolic and signaling pathways involved in them, also orchestrated by interactions of tumor, stromal and immune cells.

4.1. Tumor metabolism and its contribution to immunosuppression

Burnet and Thomas formulated the theory of cancer immunosurveillance (also called immunoediting), according to which lymphocytes would recognize and eliminate tumor cells, thus preventing tumor progression.[178,179] Nevertheless, some cancer cells are able to escape the immune response by enhancing immunosuppressive activity of immune cells. In fact, escaping immune response has been identified as one of the hallmarks of cancer.[4] Now we know that this immunosuppression is partially controlled by tumor metabolism, and also that of other cells of the TME.

High glucose uptake and lactate secretion have a major role in immunoediting inhibition. As seen above, T cells enhance glycolysis and this improves their effector function.[145] Many types of tumor cells also present a high glycolytic activity, and thereby they avidly consume glucose. As a consequence, low levels of this molecule would be available in the extracellular media for T cells consumption (Fig. 2), and then effector function would be suppressed.[180] An illustrative example is that high HK2 expression in tumor cells mitigates the transcription of the gene coding for IFN-γ, thus contributing to immune response evasion.[146] IFN-γ translation is also regulated by glycolysis through glyceraldehyde 3-phosphate dehydrogenase (GAPDH). When T cells are glucose-restricted, GAPDH becomes available to bind the 3'UTR of IFN-γ mRNA, which results in the inhibition of translation of this cytokine. A similar mechanism occurs with IL-2 (Fig. 2).[181] Furthermore, lactic acid resulting from tumor glycolysis suppresses CTL proliferation, as well as the transcription of IL-2 and IFN-γ, leading to a diminished cytotoxicity of these cells. Probably, a high extracellular level of lactic acid could block the lactic acid export, thus inhibiting further lactate production from glycolysis by T cells.[182] These observations underscore the relevance of aerobic glycolysis for the effector function of T cells. Additionally, Treg cells proliferate in response to TGF-β from tumors.[183] As a matter of fact, Treg cells are the most abundant lymphocytes in the TME. Since their energy metabolism relies on FAO and OXPHOS, they are not as vulnerable to glucose deprivation as effector T cells. In turn, Treg immunosuppressive activity contributes to overall immunosuppression within the TME.

PD-1 is an immunoinhibitory receptor expressed by chronically stimulated T cells. Ahmadzadeh et al., working with metastatic melanoma lesions, found that PD-1 is expressed by TILs at higher levels than those found in normal T cells.[183] Expression of its ligand, PD-L1, has been reported in several human tumors.[185] As PD-1/PD-L1 interaction inhibits T cell proliferation and cytokine production, it could be proposed that TME contributes to a weakened anti-tumor immune response. Different studies have shown that PD-1 expression causes a reduction of glycolysis and a switch to FAO in T cells by suppression of PI3K/Akt.[186,187] Moreover, recent results have shown that PD-L1 not only inhibits T cell glycolysis but at the same time is able to enhance this pathway in tumor cells through activation of the Akt/mTOR signaling pathway, depriving glucose availability in the TME and thus increasing even further the glycolysis inhibition in these lymphocytes.[25] Therefore, the interaction of PD-1 with its ligand PD-L1 results in an inhibition of effector T cell function (Fig. 2).

It should be emphasized that an increased tumor glycolysis is not the only way to achieve immunosuppression. We have seen that tumors avidly consume glutamine, thus depleting it from the media and affecting the immune response (Fig. 2). Moreover, many tumor cells show high levels of indoleamine-2,3-dioxygenase (IDO1) and tryptophan-2,3-dioxygenase (TDO2), two enzymes that degrade tryptophan to kynurenine. As a consequence, this amino acid is depleted from the media and effector T cells undergo apoptosis (Fig. 2).[188] Kynurenine, as mentioned in another section above, promotes invasiveness by tumor cells (Fig. 3).[165] On the other hand, expression of CD73 in some tumor cells leads to an adenosine accumulation in the extracellular media, which impairs T cell function (Fig. 2).[189] Additionally, NO production by tumor cells leads to anti-tumor immunity, whereas its production by myeloid cells promotes this anti-tumor activity.[69] Since NOS activity requires arginine as a substrate, we dare to ask whether depletion of arginine by tumor cells for the production of NO and polyamines could be the cause to the anti-tumor immunity (Fig. 2). However, combination of L-N^G-nitroarginine methyl ester (L-NAME), a NOS inhibitor, with L-arginase has been shown to reduce viability of cancer cells.[190]

In summary, not just tumor aerobic glycolysis, but also amino acid and nucleotide metabolism in tumor cells contribute to the inhibition of a proper T cell function.

4.2. Tumor and endothelial cell metabolism and its role on angiogenesis

As soon as 1971, Judah Folkman proposed that inhibiting angiogenesis could be a new and revolutionary therapy against tumor growth based on his own experimental observations from the sixties.[191] Almost 40 years later, he reviewed the available scientific information regarding a series of different angiogenesis-modulator drugs being developed for the treatment of cancer and other angiogenesis-dependent diseases, therefore reinforcing his early visionary hypothesis and now proposing that angiogenesis could be an organizing principle for drug discovery.[192] There are many factors that are related to angiogenesis (e.g. VEGF, bFGF, HIF-1α, and many others). Many published reviews have already revised this issue along the years.[193–195] Nevertheless, limitations of anti-angiogenic therapies, mainly based on the inhibition of EC activation by angiogenic factors, especially VEGF, suggested that alternative anti-angiogenic strategies might be considered.[196] The fact that metabolic reprogramming can control angiogenesis opens new horizons to treat this process under pathological conditions through a metabolic approximation and not just by targeting pro-angiogenic molecules.[197] In this section we will focus on the main metabolic features that regulate the angiogenic process, but it should be kept in mind that many other factors may interplay in this scenario.

We mentioned before that glycolysis-derived lactate plays a role in angiogenesis. Végran et al. showed that nuclear factor-κB (NF-κB) is involved in this regulation through PHD inhibition. IL-8 is a pro-angiogenic cytokine expressed by ECs. They

observed that lactate could induce IL-8 expression by these cells in a NF-κB-dependent manner. A sequence of events leading to this is proposed: lactate would be converted to pyruvate by LDH-B, which indirectly inhibits PHD2 by competition with α-ketoglutarate, with the consequent accumulation of IκB kinase (IKK), which phosphorylates inhibitor of kappa B (IκBα), thus liberating the active form of NF-κB and allowing IL-8 transcription.[97,198] Additionally, PHD inhibition enables the stabilization of HIF-1α and regulation of its target genes expression. These target genes include those coding for pro-angiogenic effectors such as VEGF and for many metabolic enzymes. HIF-1α can also indirectly induce VEGFR2 and bFGF expression. Furthermore, all this requires additionally that ECs incorporate extracellular lactate, secreted by tumor cells, through MCT1 transporters.[199] It has been shown that lactate increases the phosphorylation of Akt, thus promoting the angiogenic process.[200] VEGF plays an additional role, since it promotes fatty acid uptake by ECs, hence contributing to ECs proliferation and angiogenesis.[201] Therefore, lactate uptake by ECs would induce angiogenesis through increased IL-8, VEGF, VEGFR2 and bFGF expression and Akt phosphorylation levels (Fig. 4). Furthermore, it has been seen that extracellular lactate produced by ECs acts as a vasoactive signal for pericytes.[202] It could be possible that lactate secreted by tumor cells could also affect pericyte-mediated vasoconstriction and, thus, angiogenesis.

Moreover, recent studies have uncovered the role of nerve-endothelium interaction on angiogenesis. ECs express β_2-adrenergic receptor ($ADR\beta_2$), and its deletion leads to inhibition of angiogenesis. More specifically, $ADR\beta_2$ blockade in these cells induce a "reverse metabolic switch" towards OXPHOS, by regulation of COX10, a gene related with a cytochrome IV oxidase (Fig. 4).[203,204]

Finally, it has been recently seen that glutamine and asparagine are essential for angiogenesis.[99,100] Indeed, glutamine deprivation impairs this process, an effect rescued by the addition of asparagine and α-ketoglutarate. Consequently, inhibiting GLS1 and ASNS activities at the same time seems to be a good anti-angiogenic strategy.[99] Nevertheless, the precise mechanism of these amino acids on the angiogenic switch should be further studied.

4.3. Cancer-associated fibroblasts: important assistants for tumor invasiveness

As mentioned above, CAFs rely on enhanced glycolysis. This seems to be due to an enhanced production of ROS by cancer cells. Oxidative stress spreads from cancer cells to adjacent fibroblasts, which reduce their mitochondrial activity and increase glucose uptake, becoming more dependent on aerobic glycolysis (Figs 3 and 4).[205] In a clear example of cell cooperation within TME, CAFs secrete lactate to the media, and this lactate fuels tumor cells, which deliver it to OXPHOS, obtaining energy to sustain their high proliferative rates, in a phenomenon known as "reverse Warburg effect" (Fig. 3).[206]

Likely, this enhanced oxidative stress could induce MCT4 expression in CAFs. Moreover, co-culture of CAFs with MCF7 cells, which mostly rely on oxidative metabolism, results in an increase of MCT1 expression by these tumor cells. Thereby, lactate from CAFs would be incorporated by MCF7 cells, via a lactate shuttle between the stroma (MCT4 in CAFs) and tumor cells (MCT1 in MCF7), in a kind of tumor-feeding mechanism.[116] Something similar has also been observed in osteosarcoma.[207]

Lactate secreted by CAFs could have the same effects as those of lactate produced by tumor cells. Romero-Garcia et al. reviewed lactate contribution to the TME. From their review, the following should be highlighted: i) lactate ability to induce MMP-9, an enzyme involved in migration and invasion of cells during the angiogenic process (Fig. 4); ii) immunosuppression; iii) expression of pro-angiogenic factors; and iv) activation of ECs through MCT1.[208] Several of these processes are regulated, at least in part, by MSF expression in CAFs, a cytokine related to tumor growth.[115] However, some authors have suggested that the effects caused by extracellular acidification are specific of tumor cells.[18,120] It has been reported that lactate from cancer cells induce hyaluronic acid production by fibroblasts, contributing to tumor invasiveness (Fig. 3).[209] In addition to this, CAFs express TGF-β and SDF-1, which confer them their tumor phenotype, due to the activation of the transcriptional regulator heat shock factor 1 (HSF1), as well as pro-angiogenic features.[115,210,211] Moreover, since Treg cells proliferate in response to TGF-β from tumors, CAF-secreted TGF-β could also help the development of immunosuppression (Fig. 2).[183] It is well known that CAFs promote tumor progression and invasion, in part by secreting multiple molecules involved in ECM remodeling (Fig. 4).[212,213] Regarding angiogenesis, several available data suggest a connection between CAFs and tube formation.[214,215]

Nonetheless, lactate is not the only metabolite from CAFs that fuels tumor cells. Recent studies have shown that CAFs are able to synthetize glutamine from glutamate, aspartate and alanine, and these cells secrete this glutamine, which is used by cancer cells (Fig. 3). Again, tumor cells are not passive, but they secrete glutamate from glutaminolysis as well as the already mentioned lactate, both contributing to glutamine secretion by CAFs.[120] This interesting GS/GLS intercellular cycle within the TME deserves to be further explored. On the other hand, fatty acids are also synthetized and secreted by CAFs and taken up by breast tumor cells (Fig. 3), favoring tumor progression.[216] Furthermore, NOS-expressing CAFs support growth of breast and prostate cancer cells, suggesting the relevance of NO metabolism in these cells for tumor progression.[217]

In summary, CAFs contribute to tumor progression by fueling cancer cells, remodeling the ECM, increasing Treg proliferation and promoting angiogenesis, all in all allowing invasiveness.

4.4. Tumor-associated macrophages and tumor progression

We have seen that TAMs seem to rely on aerobic glycolysis, secreting large amounts of lactate. As a matter of fact, treatment of these cells with 2-deoxyglucose (2-DG) inhibits their pro-metastatic phenotype.[218] Interestingly, lactate from tumor cells could help by inducing aerobic glycolysis in TAMs through the Akt/mTOR pathway (Fig. 3).[135] In addition, tumor cell-derived lactate is able to induce TAM polarization by inducing *Fizz1, Mgl1* and *Mgl2* markers via HIF-1α. Additionally, VEGF and arginase 1 (Arg1) are upregulated in these cells also via HIF-1α.[219] In the first case, TAMs can be linked to angiogenesis induction. Indeed, a relationship between TAM number and tumor angiogenesis has been documented in breast cancer.[220] TAMs also produce other molecules involved in the angiogenic process, such as TNF-α, which induces MMP-9 expression, uPA, IL-1, which, through cyclooxygenase 2 (COX2), upregulates HIF-1α, increasing transcription of VEGF in turn, and CCL18.[221,222] Therefore, it is likely that TAMs help to induce tumor angiogenesis (Fig. 4). This pro-angiogenic effect of TAMs has been already seen, along with immunosuppressive features.[223]

Regarding metabolism of arginine, Arg1 has an important role in tumor progression, and participates in polyamine production, necessary for collagen synthesis, cell proliferation and tissue remodeling.[219] Indeed, some evidence hint that TAMs could contribute to tumor invasion by secreting MMPs.[224] There is some controversy regarding the presence of iNOS expression in TAMs. iNOS is an enzyme that produces NO from arginine. This enzyme is present in M1 macrophages whereas is absent in M2 macrophages.[225] Regarding metabolic features of these cells, iNOS is able to block OXPHOS while upregulating the glycolytic rate, and therefore iNOS expression corresponds with M1 and TAM metabolic profiles.[226] Some authors have found iNOS expression in TAMs while others could not.[227,228] On the other hand, TAMs could have a role in immunosuppression, since depleting extracellular arginine by Arg1 activity would deprive T cells of this amino acid, affecting their proliferation.[229] Moreover, TAMs express high levels of IDO, producing kynurenine (Fig. 3), and this tryptophan degradation impairs T cell function.[230] These data reflect the immunosuppressive capacity of TAMs (Fig. 2), and iNOS has an immunosuppressive (as well as anti-angiogenic) effect.[14] Therefore, additional experiments should be performed in order to confirm the involvement of iNOS in these cells.

Furthermore, these macrophages are unable to produce IL-12, a cytokine required to activate the anti-tumor responses mediated by NK cells, Th1 cells and CTLs. Instead, they produce IL-10, inducing Th2 polarization, and these Th2 cells secrete IL-4, promoting M2 polarization to TAMs in a positive-feedback cycle.[122] Th2 cells release anti-inflammatory cytokines, so they do not contribute to the anti-tumor immune response. IL-10 secreted by TAMs also increases the number of Treg cells present in epithelial ovarian cancer (Fig. 2).[231] It has been demonstrated recently that IL-10 inhibits mTOR activation in macrophages, thus leading to a reduction in the glycolytic pathway and ROS liberation from damaged mitochondria.[232,233] Since mTOR inhibition promotes Treg cell differentiation, a relationship between IL-10 from TAMs and mTOR in tumor progression may be established.[148]

In addition, as tumor cells, TAMs also express PD-L1, contributing to immunosuppression (Fig. 2).[234] Lactate secreted by cancer cells is able to increase IL-23 secretion by TAMs, a tumor-promoting cytokine involved in the generation of Th17 cells, thus contributing to tumor progression.[235]

Fatty acid and glutamine metabolism in TAMs are also important for tumor progression. For example, an elevated FA biosynthesis, uptake or storage contributes to the pro-tumorigenic profile of these cells.[225] On the other hand, TAMs show high levels of GS expression, thus liberating glutamine to the media for feeding tumor cells and contributing to nitrogen metabolism in these cells, as CAFs do (Fig. 3).[138,219]

All these facts indicate that TAMs can help tumor cells to evade the immune response, to trigger tumor angiogenesis and to promote invasiveness.

4.5. Other examples of "friendly neighbors" of tumors

Not mentioned above, ECs are able to help tumor cells within the TME. For example, they can extrude mitochondria to tumor cells through tunneling nanotubes and thus they can acquire resistance to chemotherapy (Fig. 3).[236] However, the already mentioned stromal cells are not the only ones able to help tumors to grow. Depending on the type of cancer, there can be other cells that feed tumor cells. They could be called "friendly neighbors", as in the title of a comment regarding a letter which described the alanine release from pancreatic stellate cells to tumor cells in the pancreas.[237,238] Some mesenchymal stromal cells have been shown to take up cystine and convert it into cysteine, which is released and taken up by tumor cells from chronic lymphocytic leukemia (CLL). These cancer cells use this cysteine for glutathione (GSH) synthesis, involved in cell survival and resistance to drug cytotoxicity.[239] As CAFs and TAMs do, adipocytes in pancreatic cancer synthetize and secrete glutamine to the media and thus they can feed tumor cells.[240] But that is not all: it has been seen in several types of cancer that adipocytes release fatty acids that are used as fuels by tumor cells, thus contributing to invasiveness, as in the case of CAFs.[241–244] Moreover, NO-mediated S-nitrosylation triggers adipocyte formation, thus providing tumor cells a source of fatty acids.[245] In addition, adipocytes also secrete arginine that are used by tumor cells to NO synthesis, and the resulting citrulline is taken up by adipocytes in a cross-talk between both cells (Fig. 3).[190]

5. HOST METABOLISM ALTERATIONS AFTER TUMOR DEVELOPMENT

We have already revised some features and implications of the metabolism of the cells within the TME. However, it should not be forgotten that this TME is just a small

part of the organism bearing the tumor. Tumor angiogenesis developed by ECs in the TME allows the secretion of several soluble factors to the circulation, which leads to pathological endocrine effects and an interaction of this microenvironment with the rest of the tissues. So, we cannot just talk about a TME, but a tumor macroenvironment should be as well (or even more importantly) considered, since cancer-associated systemic syndromes develop in this disease.[246]

The concept of the "systemic effect" was firstly proposed by Shapot. He affirmed that all malignant tumors alter host homeostasis and metabolism even in the absence of metastasis, whereas benign tumors do not share this property.[247] He distinguished between two manifestations of this systemic effect: i) the alteration of the host metabolism by competence of the tumor with host tissues, and ii) a dysregulation of endocrine gland activities and, therefore, a diminished sensitivity to hormones.[247] Recently, the concept of solid tumors as systemic metabolic dictators has been proposed.[248]

The most classical feature of tumors in the context of their interaction with the host is the concept of tumors acting as "nitrogen traps". As early as 1889, Müller observed a negative nitrogen balance in patients with malignant tumors.[referred in 249] Nevertheless, the concept of nitrogen trap was firstly demonstrated by Mider.[250] Moreover, because glutamine is the most abundant amino acid in blood, some authors consider tumors as "glutamine traps".[251] Early results obtained by our group in Ehrlich ascites tumors suggested that tumors elicit a specific response from the host tissues, so that the whole organism contributes to supply glutamine to the tumor.[33] Indeed, glutamine content in the host decreases in fast growing tumors due to a flux of glutamine from the host to the tumor, low or null GS activity in the tumor and faster transport of glutamine through the plasma membrane of tumor cells in comparison with non-tumor cells (Fig. 5).[12,252] There is the exception of some tumors that present a GS upregulation as an adaptation to glutamine depletion, a feature that is not specific to tumor cells.[253,254] In spite of the importance of glutamine for tumors, changes in concentrations of other amino acids are also observed in plasma after tumor transplantation due to the host-tumor interaction.[255]

This nitrogen trap may have other effects in the organism. For example, it has been seen that tumors intercept uridine from lymphoid organs, thus inhibiting RNA synthesis, and DNA synthesis is suppressed in the spleen of tumor-bearing mice.[256,257] Due to the avid host glutamine consumption by the tumor, concentration of glutathione in natural killer cells diminishes, with the consequent loss of activity of these cells.[258] All these data support that tumors acting as glutamine traps also compromise the immune system response and, therefore, there is an immunosuppression helped by the alteration of nitrogen metabolism in the host (Fig. 5). Some authors have observed that an oral supplement of glutamine in the diet can have benefits in tumor-bearing animals and cancer patients, although a consensus about this has not been achieved.[259,260]

But tumors not only take nitrogen from the diet. They are also able to take it from host tissues with the consequent body weight loss.[250,261] However, tumor grows to a

lesser extent when there is no nitrogen available from the diet.[262] This loss in body weight leads to cancer-associated cachexia.[263] Nitrogen from host tissues proceeds from protein catabolism, stimulated by an upregulated production of adrenocortical hormones (ACH) resulted from a dysregulation of the endocrine system (Fig. 5).[264] This dysregulation can lead to other harmful effects in the organism, such us thrombosis and immunosuppression.[265,266] Now we know that this upregulation of glucocorticoid production is caused by IL-6 secretion from the tumor through inhibition of some hepatic functions such as ketogenesis (Fig. 5).[267] As a matter of fact, inhibition of IL-6 diminishes tumor growth and cachexia.[268]

It has been seen that IL-6 from lung adenocarcinoma is able to inhibit another characteristic of liver metabolome, such as hepatic insulin signaling.[269] This insulin resistance contributes to protein catabolism and induction of glucogenolisis and gluconeogenesis (Fig. 5). Indeed, gluconeogenesis is induced by glucocorticoids after tumor transplantation, and lower levels of glycogen are found in the liver of tumor-bearing animals.[249,270] Glucose can be synthetized from gluconeogenic amino acids. These amino acids include glutamine, which is used mainly in kidneys, and alanine, used almost exclusively by the liver.[271] A significant part of this gluconeogenic glutamine comes from catabolism of muscle proteins, which reflects the correlation between cachexia and gluconeogenesis.[272] Very recently, a study of plasma metabolome from breast cancer patients revealed a positive correlation between lactate, pyruvate and alanine levels, and a negative correlation of pyruvate and alanine with glucose.[273] This corresponds with the Cori cycle, an inter-system cycle active in tumor patients: lactate released from cancer cells, but also from muscles, goes to the liver, as well as alanine from muscle, and these metabolites are used in gluconeogenesis in that organ, increasing the glucose available for cancer cells and their stroma, and thus enhancing tumor malignancy and associated body weight loss (Fig. 5).[274] Moreover, the use of amino acids for gluconeogenesis limits the protein synthesis in the host, contributing to vital organs dystrophy.[249] Indeed, a low amount of membrane-bound ribosomes and a defect of the small subunit of ribosomes in muscle were found in tumor-bearing animals.[275,276]

Due to the Warburg effect, many tumors depend on aerobic glycolysis. For that reason, tumors can also be considered as "glucose traps".[249] The consequent decrease in glucose levels due to its consumption by the tumor is, in part, responsible for the up-regulated glycogenolysis and gluconeogenesis. But that is not all. Administration of additional glucose inhibits fatty acid mobilization in the host, showing a modulation of fatty acid metabolism due to glucose depletion caused by the tumor.[277] As a matter of fact, lipid catabolism in adipocytes promotes cancer-associated cachexia in tumor-bearing mice.[278] This mobilization of fatty acids could also be associated with fatty acid synthesis in tumors, as serum levels of fatty acids were found to be lower in tumor-bearing mice as compared to the controls (Fig. 5).[279]

Interestingly, supplementation of arginine in the diet inhibits body weight loss and diminishes tumor growth as well as nitrogen trapped by the tumor. On the one hand, increased leucine oxidation due to additional, available arginine leads to a decrease in protein catabolism.[280] On the other hand, arginine is able to activate the immune system, with the consequent reduction of tumor growth.[281] Nowadays we know the importance of arginine in T cells activity.[152] We would like to highlight the use of arginine for polyamine synthesis, a process enhanced in tumors that could be hence responsible for immunosuppression by depleting extracellular arginine (Fig. 5).

Other amino acids can be taken up by tumors from host tissues. A flux of several essential amino acids, such as valine, leucine, isoleucine, phenylalanine, lysine and arginine, as well as the sulfur amino acid methionine, was observed in Ehrlich carcinoma-bearing mice.[255] Regarding methionine flux, this could be explained by the active polyamine biosynthesis in the tumor, also demonstrated by the observation of a net flux of ornithine from host to tumor and an increase in ODC activity in the seventh day after tumor transplantation in the same animal model (Fig. 5).[282] Moreover, tumors can take cysteine and incorporate it through CD44 in order to synthetize glutathione. It has been seen that CD44 interacts with PKM2, increasing the Warburg effect. Therefore, inhibition of this cell marker leads to an increased glucose oxidation and reduced glutathione levels in tumor cells, enhancing the oxidative damage in these cells.[283]

In addition of inducing protein catabolism in the host and hence acquiring amino acids, Ras-mutant tumor cells are able to incorporate extracellular proteins (mostly serum albumin) by macropinocytosis, and to obtain amino acids from their lysosomal degradation for sustaining cell proliferation even in the absent of extracellular glutamine.[284,285] Indeed, Holm et al observed that the amount of nitrogen excreted in colorectal cancer was 10-fold higher than the equivalent amino acid uptake, pointing out the possible incorporation of extracellular proteins.[286] PIKfyve has been demonstrated to promote recovery and redistribution of nutrients from vacuoles after lysosomal degradation of engulfed proteins, thus supporting Ras-mutant cell proliferation.[287] On the other hand, an input of amino acids results in mTORC1 activation, which inhibits lysosomal catabolism of extracellular proteins.[288] Besides, oncogene Ras does not only induce macropinocytosis of extracellular proteins, but it also induces lipid scavenging, thus conferring resistance to inhibition of stearoyl-CoA desaturase 1 (SCD1), a key enzyme in fatty acid metabolism.[289] Novel therapeutic strategies are emerging based on these discoveries. For example, drug conjugation with albumin (e.g. paclitaxel) increases intratumoral drug concentration and enhances anti-tumoral activity.[290,291] mTORC1 inhibitors have sometimes failed in suppressing tumor growth. Combination of mTORC1 inhibitors with blockade of extracellular proteins macropinocytosis or PIKfyve inhibitors could be a promising combined strategy for Ras-mutant tumors.[287,288]

In summary, host-tumor interactions and the presence of extracellular substrates are of great importance for tumor progression, and metabolism plays an essential role. Despite the relevance of host metabolism in tumors, just a few studies have been performed in the last years, and the vast majority of research regarding this issue is previous to the present century. Therefore, more research would be necessary in order to improve treatment for cancer patients taking into account the whole organism homeostasis.

6. TARGETING METABOLISM OF TUMOR MICROENVIRONMENT CELLS FOR CANCER THERAPY

The "re-discovery" of the Warburg effect and increased glutaminolysis and the identification of tumor metabolism reprogramming as a hallmark of cancer renewed the interest in cancer metabolism after decades of oversight and has led to a renewed interest in targeting tumor metabolism in the last two decades. Many compounds targeting cancer metabolism have been tested *in vitro*, *in vivo* and in clinical trials. These compounds include glycolysis inhibitors like 2-DG, lonidamine, 3-bromopyruvate and dichloroacetate and inhibitors of GLS such as 968, BPTES and other glutamine analogues, including DON, acivicin and azaserine, among many others.[7,11-13] However, the search for anti-glutamine cancer therapies, despite good results in *in vivo* models, was soon forgotten.[292] A renewed interest in these agents has been recently triggered by the observation that GLS inhibitors may help to overcome acquired resistance to anti-tumor drugs in ovarian and non-small-cell lung cancer.[293–296] Inhibiting polyamine metabolism has also been shown to decrease tumor growth, and its targeting is considered of great relevance for cancer therapy.[85,297] Additionally, treatment using asparaginase has been proved to be useful against leukemia. Moreover, this enzyme has a well-known immunosuppressor role, that can be explained by an almost undetectable ASNS activity in lymphoid tissues and the glutaminase activity presented in most asparaginases.[298-300] Therefore, since treatment with asparagine inhibits T cell activation as well as cytokine production and proper function of M1 macrophages, it should be taken into account that targeting asparagine metabolism in tumors could also affect the immune system.[301,302]

Furthermore, the concept of "oncometabolites" has opened a new window for tumor treatment. We could define the term oncometabolite as a molecule from normal metabolism that is able to allow tumor progression through its accumulation due to a metabolic dysregulation. The best and first known oncometabolite is 2-HG, which causes changes in gene function in tumors by epigenetic regulation.[43] One of the consequences of the accumulation of 2-HG to limit the production of chemokines CXCL9 and CXCL10, so preventing $CD8^+$ T cell recruitment to the tumor, for example.[303] In the last years, efforts to inhibit the newly gained function of the mutant IDH enzymes (IDH1 and IDH2) have led to the development of IDH inhibitors which

are already in clinical trials.[304–306] Other molecules are also considered as oncometabolites, and their targeting should also be researched.[307,308]

However, in the last years alternatives have emerged with the new understanding of the complex metabolic interactions within the TME. As we have shown above, overall TME metabolic features are sometimes determined by cytokines or pro-angiogenic factors production. In fact, chemoresistance is sometimes enhanced due to interactions with stromal cells and components of the ECM.[309] On the other hand, it is known that non-tumor cells are genetically more stable than tumor cells, and thus it is less likely that these cells could develop adaptive mutations to treatments.[224] Therefore, targeting metabolism of TME stromal cells, instead of tumor cell metabolism or in addition to it, could be a promising strategy against tumor progression.

Since metabolism and angiogenesis are related, it could be expected that metabolic modulators were also able to affect different steps of the angiogenic process. Among other examples, 3-bromopyruvate, an inhibitor of hexoquinase, and α-cyano-4-hydroxycinnamic acid (CHC), which blocks MCT lactate transporter, inhibit angiogenesis in HUVEC.[310] 2-DG, the most well-known glycolytic inhibitor, inhibits angiogenesis *in vitro* and *in vivo*.[311] The glycolytic pathway is not the only possible target. For instance, acivicin, a glutamine analogue, disrupts angiogenesis *in vivo*, and chloroquine, a GDH inhibitor, enhances the anti-angiogenic effect of sunitinib.[312,313] In addition, some statins, HMG-CoA reductase inhibitors that affect metabolism of cholesterol, and DFMO, an inhibitor of ODC, involved in polyamine metabolism, are capable of suppressing the angiogenic process.[314-316]

Recently, three articles simultaneously published in *Cell Reports* have demonstrated that the induction of metabolic symbiosis could be responsible for acquired resistance to anti-angiogenic drugs.[317-319] Treatment with inhibitors of angiogenesis, including sunitinib, may give rise to an extensive vascular collapse that will produce hypoxic and normoxic regions in the tumor. In the hypoxic cancer cells, HIF-1α induction will upregulate GLUT1 and MCT4, leading to high levels of lactate secretion. This lactate will be imported by the normoxic cancer cells, which express the lactate transporter MCT1, and catabolized with consequent induction of mTOR signaling to promote tumor metabolism. In this way, normoxic cancer cells save glucose for the hypoxic cells and use the lactate produced by hypoxic cells in conjunction with glutamine.[317] Targeting metabolic symbiosis may therefore be a new strategy to overcome the resistance development to anti-angiogenic therapy in patients.

Targeting EC metabolism could be, as well, a way to inhibit tumor angiogenesis.[197] Inhibition of PFKFB3 and pharmacological blockade of MCT1 disrupt angiogenesis *in vitro* and *in vivo*, and LDH-A inhibition impairs proliferation of pulmonary microvascular ECs.[93,199,320] Indeed, taking EC metabolism as a target for modulating pathological angiogenesis may improve chemotherapy, as seen for a PFKFB3 inhibitor, 3-PO, which impairs metastasis without affecting proliferation of tumor cells.[321] After uncovering the importance of fatty acid metabolism in ECs, targeting fatty acid

synthesis and oxidation is emerging as a novel therapeutic approach to inhibit EC metabolism and angiogenesis.[106,322] Furthermore, etomoxir, a CPT1a inhibitor, represses angiogenesis.[106] Glutamine and asparagine metabolism are also emerging targets for inhibition of the angiogenesis process.[99,100]

Many anti-angiogenic compounds are available and already approved for their use in patients.[192,323] Moreover, a combinatory strategy is also being explored, since sometimes anti-angiogenic therapy may be not enough to treat tumors.[5] This anti-angiogenic therapy could result in i) the recovery of the normal perfusion in tissue, with the consequent reduction in hypoxia and an improvement of the immunosupportive immune system, ii) no change or iii) excessive pruning of the vasculature, with a decrease in blood flow and an increase in hypoxia.[324] Therefore, its combination with metabolic modulators or with immunotherapy could improve the treatment.[324–326]

The use of inhibitors of lactate transport and production could be a good strategy to target the reverse Warburg effect in stromal cells, and not just lactate metabolism in tumor cells. An inhibitor of MCT1 (AZD3965) is already in phase I trials to this aim.[327] Similarly, metformin can also be used to target stromal cells in addition of tumor cells. It has been shown that this drug can block lipid accumulation in ovarian cancer cells adjacent to adipocytes, and reverse the malignant phenotype of CAFs by restoring caveolin-1 expression in these cells.[328,329] Other possibilities are targeting GS in CAFs, as well as GLS in tumor cells, in order to avoid glutamine transfer from CAFs to cancer cells.[120] Other suggested therapies based on targeting stromal cell metabolism (such as CAFs and CAAs) are collected in the bibliography.[330]

The denominated checkpoint blockade therapy using antibodies against PD-L1 has emerged as a strategy to restore glucose in the TME and recover T cell effector function in order to suppress tumor progression.[25] Since tumor and T cells share many metabolic features, targeting their metabolism can have undesired effects. For example, administration of mTOR inhibitors can either promote effector T cells or inhibit them. Furthermore, blocking glycolysis could affect T cell metabolism and lead to a poor prognosis of cancer. However, the use of glycolytic inhibitors before the induction of an immune response may allow T cells to enter a TME with higher glucose concentration, favoring a proper anti-tumor immune response.[15] Combining an anti-metabolic strategy with a checkpoint blockade therapy could improve the T cell function and cancer prognosis. For example, it has been reported that targeting CD73 in tumors enhances the efficacy of anti-PD-1 and anti-CTLA-4 treatments.[331]

Anti-tumor T cell function can be also partially recovered by inhibiting Arg1 with tadalafil.[14] Inhibitors of IDO have been proposed to restore T cell proliferation and cytokine production, and dimethylfumarate (DMF), an anti-angiogenic compound, is able to inhibit IDO activity in human immune cells.[17,332,333] Moreover, very recently an inhibitor of IDO, erianin, has also been shown to inhibit tumor angiogenesis.[334]

In summary, targeting stromal cell metabolism and development of immunotherapy with metabolism as a target may improve cancer therapies by inhibiting angiogenesis and recovering anti-tumor immune response, leading to tumor regression. Several compounds able to modulate metabolic features with proved anti-tumor activity are collected in Table 1. However, it is always important to be careful with secondary effects and to make sure that normal metabolism is not affected by the treatment. Further research will be necessary to progress on cancer treatment via inhibition of the TME metabolism.

7. CONCLUDING REMARKS AND OUTSTANDING QUESTIONS

In this review we have tried to explore metabolism within the TME and how it affects tumor growth and progression. Four major kinds of cells have been analyzed: ECs, TILs, CAFs and TAMs, apart from tumor cells. Summarizing, all these cells rely mainly on aerobic glycolysis with the exception of Treg cells, which mainly depend on an oxidative metabolism. Lactate production by tumor cells would contribute to promote tumor angiogenesis via NF-κB and HIF-1α stabilization. TAMs and CAFs also collaborate by secreting pro-angiogenic factors. During tumor progression a process termed immunosuppression occurs, by which T cells are unable to exert a proper anti-tumor immune response. Tumor cells, by glucose competition and lactate secretion, as well as other metabolic features of these and other cells, are responsible for this. PD-1/PD-L1 interaction is also a way to immunosuppression, in which tumor cells, T cells and TAMs are implicated. CAFs also fuel tumor cells by a phenomenon called reverse Warburg effect and by glutamine synthesis and secretion, along with TAMs and CAAs.

Although in this review we have focused on the changes regarding metabolism in the TME, metabolism is considered a complex and dynamic network able to adapt in response to shifts and metabolic demands.[330] Therefore, cancer metabolic reprogramming is just an example of the flexibility and adaptability of metabolism. Circadian rhythms, hypoxia, exercise, hibernation period and many other factors are able to modulate gene expression and metabolic features of healthy cells.[335-338] The lactate shuttle between tumor cells and other cells of their microenvironment is also present in healthy tissues, such as muscle and brain.[339-342] Moreover, it has been recently demonstrated that there are also changes in metabolism during developmental progression and not just during differentiation, and a loss of metabolic flexibility could lead to pathologies associated to metabolic syndrome.[343] Actually, this metabolic flexibility is not only found in animals, but in all organisms. Plants, for example, are able to modify their metabolism in response to environmental stress.[344,345] Due to this metabolic flexibility, tumors can modulate the metabolism of the tissues in the so-called systemic effect. Therefore, not only metabolism of the sole TME, but also the changes in the metabolism of the whole organism triggered by the tumor should be studied.

In conclusion, although it is obvious and well-documented that there is a metabolic switch during tumor progression, these kinds of changes also take place in healthy tissues as a normal process or under particular situations and they should not be considered as surprising. All in all, cancer metabolic reprogramming ought to be studied as an ordinary and expected feature of metabolism. Regarding possible therapies, targeting the metabolic features of the different cells of the TME, or putting the target in the angiogenic process or the immune system, will allow us to design new strategies to fight cancer in combination with classical metabolic approaches.

We could take into consideration the next remarkable aspects: i) aerobic glycolysis is upregulated in different cells of the TME, except for Treg cells; ii) tumor cells should be classified as oxidative and glycolytic ones, even within the same tumor; iii) due to different metabolic modulations, cells of the TME help to tumor progression, affecting invasiveness, angiogenesis and immunosuppression; iv) tumor macroenvironment should not be rotten in oblivion, and more research should be performed in order to improve treatments; v) metabolism regulates and is linked to many other physiological characteristics, being part of an interconnected network; vi) the concept of metabolic switch is not specific of cancer, but an example of the global flexibility of metabolism.

Finally, we bring together some questions that remain up in the air waiting for being elucidated: i) Is there any glucose competition between tumor and ECs? And between tumor, CAFs and TAMs? ii) What is the exact mechanism by which lactate undermines T cells glycolytic metabolism? iii) What is the exact role of arginine in the immune system? iv) Which metabolic features characterize TAMCs and tumor-associated pericytes? What is their role in tumor progression? Further investigation will be needed to solve these inquiries.

NOTES ADDED IN PROOF

During the revision period of this article a study showing an interaction between metabolic reprogramming and transcriptional regulation has been published. Dasgupta et al. have shown that the metabolic enzyme 6-phosphofructo-2-kinase/fructose-2,6-biphosphatase 4 (PFKFB4) regulates transcriptional programming by activating the oncogenic steroid receptor coactivator-3 (SRC-3) through its phosphorylation at serine 857. An active glucose metabolism allows this phosphorylation, which leads to upregulation of some of the key enzymes of the pentose phosphate pathway (PPP). This activation of purine metabolism is essential for tumor growth and metastasis in breast cancer models, since ablation of SRC-3 or PFKFB4 leads to a decrease in cell growth and the metastatic progression of the disease.[431] Another enzyme of the same family, PFKFB3, was shown to be involved in angiogenesis.[93] Hence, we would like to remark the importance of metabolism in the development of diseases such as cancer and angiogenic-dependent pathologies through different mechanisms.

We have also become aware of the approval by FDA of enasidenib for the treatment of oncologic patients with tumor *IDH2* gene mutations.[432]

References

1. Warburg O. The metabolism of carcinoma cells. *J Cancer Res* 1925;9:148–163.

2. Warburg O. On the origin of cancer cells. *Science* 1956;123(3191):309–314.

3. Gatenby RA, Gillies RJ. Why do cancers have high aerobic glycolysis? *Nat Rev Cancer* 2004;4:891–899.

4. Hanahan D, Weinberg RA. Hallmarks of cancer: The next generation. *Cell* 2011;144:646–674.

5. Quesada AR, Medina MA, Alba E. Playing only one instrument may be not enough: Limitations and future of the antiangiogenic treatment of cancer. *BioEssays* 2007;29:1159–1168.

6. Nyberg P, Salo T, Kalluri R. Tumor microenvironment and angiogenesis. *Front Biosci* 2008;13:6537–6553.

7. López-Lázaro M. A new view of carcinogenesis and an alternative approach to cancer therapy. *Mol Med* 2010;16(3–4):144–153.

8. Moreno-Sánchez R, Rodríguez-Enríquez S, Marín-Hernández A, Saavedra E. Energy metabolism in tumor cells. *FEBS J* 2007;274:1393–1418.

9. Pelicano H, Martin DS, Xu RH, Huang P. Glycolysis inhibition for anticancer treatment. *Oncogene* 2006;25:4633–4646.

10. Schulze A, Harris AL. How cancer metabolism is tuned for proliferation and vulnerable to disruption. *Nature* 2012;491(7424):364–373.

11. Vander Heiden MG. Targeting cancer metabolism: a therapeutic window opens. *Nat Rev Drug Discov* 2011;10:671–684.

12. Medina MA, Sánchez-Jiménez F, Márquez J, Quesada AR, Núñez de Castro I. Relevance of glutamine metabolism to tumor cell growth. *Mol Cell Biochem* 1992;113:1–15.

13. Katt WP, Cerione RA. Glutaminase regulation in cancer cells: a druggable chain of events. *Drug Discov Today* 2013;19(4):450–457.

14. Buqué A, Bloy N, Aranda F, et al. Trial Watch-Small molecules targeting the immunological tumor microenvironment for cancer therapy. *Oncoimmunology*

2016;5(6):e1149674.

15. Chang CH, Pearce EL. Emerging concepts of T cell metabolism as a target of immunotherapy. *Nat Immunol* 2016;17(4):364–368.

16. Noy R, Pollard JW. Tumor-Associated Macrophages: From Mechanisms to Therapy. *Immunity* 2014;41(1):49–61.

17. Mockler MB, Conroy MJ, Lysaght J. Targeting T cell immunometabolism for cancer immunotherapy; understanding the impact of the tumor microenvironment. *Front Oncol* 2014;4:107.

18. Pavlova NN, Thompson CB. The emerging hallmarks of cancer metabolism. *Cell Metab* 2016;23:27–47.

19. Dell' Antone P. Energy metabolism in cancer cells: How to explain the Warburg and Crabtree effects? *Med Hypotheses* 2012;79:388–392.

20. Arora R, Schmitt D, Karanam B, Tan M, Yates C, Dean-Colomb W. Inhibition of the Warburg effect with a natural compound reveals a novel measurement for determining the metastatic potential of breast cancers. *Oncotarget* 2015;6(2):662–678.

21. Guppy M, Leedman P, Zu X, Russell V. Contribution by different fuels and metabolic pathways to the total ATP turnover of proliferating MCF-7 breast cancer cells. *Biochem J* 2002;364:309–315.

22. Chen Z, Odstrcil E a, Tu BP, McKnight SL. Restriction of DNA replication to the reductive phase of the metabolic cycle protects genome integrity. *Science* 2007;316:1916–1919.

23. Lunt SY, Vander Heiden MG. Aerobic glycolysis: Meeting the metabolic requirements of cell proliferation. *Annu Rev Cell Dev Biol* 2011;27:441–464.

24. Vander Heiden MG, Cantley L, Thompson C. Understanding the Warburg effect: The metabolic requiremetns of cell proliferation. *Science* 2009;324(5930):1029–1033.

25. Chang CH, Qiu J, O'Sullivan D, et al. Metabolic competition in the tumor microenvironment is a driver of cancer progression. *Cell* 2015;162:1–13.

26. Laing RE, Nair-Gill E, Witte ON, Radu CG. Visualizing cancer and immune cell function with metabolic positron emission tomography. *Curr Opin Genet Dev* 2010;20(1):100–105.

27. Reitzer LJ, Wice BM, Kennell D. Evidence that glutamine, not sugar, is the major energy source for cultured HeLa cells. *J Biol Chem* 1979;254(8):2669–

2676.

28. Gentric G, Mieulet V, Mechta-Grigoriou F. Heterogeneity in cancer metabolism: new concepts in an old field. *Antioxid Redox Signal* 2016;0(0):1–24.

29. Pérez-escuredo J, Hée VF Van, Sboarina M, et al. Monocarboxylate transporters in the brain and in cancer. *Biochmica Biophys Acta* 2016;1863:2481–2497.

30. Hui S, Ghergurovich JM, Morscher RJ, et al. Glucose feeds the TCA cycle via circulating lactate. *Nature* 2017;551:115–118.

31. Lu W, Pelicano H, Huang P. Cancer metabolism: Is glutamine sweeter than glucose? *Cancer Cell* 2010;18:199–200.

32. Carrascosa JM, Martínez P, Núñez de Castro I. Nitrogen movement between host and tumor in mice inoculated with Ehrlich ascitic tumor cells. *Cancer Res* 1984;44:3831–3835.

33. Quesada AR, Medina MA, Márquez J, Sánchez-Jiménez FM, Núñez de Castro I. Contribution by host tissues to circulating glutamine in mice inoculated with Ehrlich ascites tumor cells. *Cancer Res* 1988;48:1551–1553.

34. Segura JA, Medina MA, Alonso FJ, Sanchez-Jimenez F, Núñez de Castro I. Glycolysis and glutaminolysis in perifused Ehrlich ascites tumour cells. *Cell Biochem Funct* 1989;7(1):7–10.

35. DeBerardinis RJ, Cheng T. Q's next: The diverse functions of glutamine in metabolism, cell biology and cancer. *Oncogene* 2010;29(3):313–324.

36. Filipp F V, Ratnikov B, De Ingeniis J, Smith JW, Osterman AL, Scott DA. Glutamine-fueled mitochondrial metabolism is decoupled from glycolysis in melanoma. *Pigment Cell Melanoma Res* 2012;25(6):732–739.

37. DeBerardinis RJ, Mancuso A, Daikhin E, et al. Beyond aerobic glycolysis: Transformed cells can engage in glutamine metabolism that exceeds the requirement for protein and nucleotide synthesis. *Proc Natl Acad Sci* 2007;104(49):19345–19350.

38. Spinelli JB, Yoon H, Ringel AE, Jeanfavre S, Clish CB, Haigis MC. Metabolic recycling of ammonia via glutamate dehydrogenase supports breast cancer biomass. *Science* 2017.

39. Floor SL, Dumont JE, Maenhaut C, Raspe E. Hallmarks of cancer: Of all cancer cells, all the time? *Trends Mol Med* 2012;18(9):509–515.

40. Elia I, Schmieder R, Christen S, Fendt S-M. Organ-specific cancer metabolism and its potential for therapy. *Handb Exp Pharmacol* 2016;(233):321–353.

41. Eason K, Sadanandam A. Molecular or metabolic reprograming: what triggers tumor subtypes? *Cancer Res* 2016;76(18):5195–5200.

42. Ruiz-Pérez MV, Sánchez-Jimenez F, Alonso FJ, Segura JA, Márquez J, Medina MA. Glutamine, glucose and other fuels for cancer. *Curr Pharm Des* 2014;20(15):2557–2579.

43. Yun J, Johnson JL, Hanigan CL, Locasale JW. Interactions between epigenetics and metabolism in cancers. *Front Oncol* 2012;2:163.

44. Vasudevan D, Bovee RC, Thomas DD. Nitric oxide, the new architect of epigenetic landscapes. *Nitric Oxide* 2016;59:54–62.

45. Bloch-Frankenthal L, Langan J, Morris HP, Weinhouse S. Fatty acid oxidation and ketogenesis in transplantable liver tumors. *Cancer Res* 1965;25:732–736.

46. Caro P, Kishan AU, Norberg E, et al. Metabolic signatures uncover distinct targets in molecular subsets of diffuse large B cell lymphoma. *Cancer Cell* 2012;22:547–560.

47. Fields ALA, Wolman SL, Cheema-Dhadli S, Morris HP, Halperin ML. Regulation of energy metabolism in Morris hepatoma 7777 and 7800. *Cancer Res* 1981;41:2762–2766.

48. Tisdale MJ, Brennan RA. Metabolic substrate utilization by a tumour cell line which induces cachexia in vivo. *Br J Cancer* 1986;54:601–606.

49. Beloribi-Djefaflia S, Vasseur S, Guillaumond F. Lipid metabolic reprogramming in cancer cells. *Oncogenesis* 2016;5:e189.

50. Shurbaji MS, Kalbfleisch JH, Thurmond TS. Immunohistochemical detection of a fatty acid synthase (OA-519) as a predictor of progression of prostate cancer. *Hum Pathol* 1996;27(9):917–921.

51. Rashid A, Pizer ES, Moga M, et al. Elevated expression of fatty acid synthase and fatty acid synthetic activity in colorectal neoplasia. *Am J Pathol* 1997;150(1):201–208.

52. Wang Y, Kuhajda FP, Li JN, et al. Fatty acid synthase (FAS) expression in human breast cancer cell culture supernatants and in breast cancer patients. *Cancer Lett* 2001;167:99–104.

53. Santos CR, Schulze A. Lipid metabolism in cancer. *FEBS J* 2012;279:2610–2623.

54. Samudio I, Harmancey R, Fiegl M, et al. Pharmacologic inhibition of fatty acid oxidation sensitizes human leukemia cells to apoptosis induction. *J Clin Invest*

2010;120(1):142–156.

55. Uray IP, Liang Y, Hyder SM. Estradiol down-regulates CD36 expression in human breast cancer cells. *Cancer Lett* 2004;207(1):101–107.

56. Pascual G, Avgustinova A, Mejetta S, et al. Targeting metastasis stem cells through the fatty acid receptor CD36. *Nature* 2016;541:41–45.

57. Zhang J, Fan J, Venneti S, et al. Asparagine plays a critical role in regulating cellular adaptation to glutamine depletion. *Mol Cell* 2014;56:205–218.

58. Broome J. Evidence that the L-asparaginase of guinea pig serum is responsible for its antilymphoma effects. *J Exp Med* 1963;118:99–120.

59. Haskell C, Canellos G. L-Asparaginase resistance in human leukemia - Asparagine synthetase. *Biochem Pharmacol* 1969;18:2578–2580.

60. Aslanian AM, Fletcher BS, Kilberg MS. Asparagine synthetase expression alone is sufficient to induce L-asparaginase resistance in MOLT-4 human leukaemia cells. *Biochem J* 2001;357:321–328.

61. Balasubramanian MN, Butterworth EA, Kilberg MS. Asparagine synthetase: regulation by cell stress and involvement in tumor biology. *Am J Physiol - Endocrinol Metab* 2013;304:E789–799.

62. Dufour E, Gay F, Aguera K, Scoazec J. Pancreatic tumor sensitivity to plasma L-asparagine starvation. *Pancreas* 2012;41(6):940–948.

63. Karpel-massler G, Ramani D, Shu C, et al. Metabolic reprogramming of glioblastoma cells by L-asparaginase sensitizes for apoptosis in vitro and in vivo. *Oncotarget* 2017;7(23):33512–33528.

64. Amelio I, Cutruzzolá F, Antonov A, Agostini M, Melino G. Serine and glycine metabolism in cancer. *Trends Biochem Sci* 2014;39(4):191–198.

65. Farber S, Diamond L., Mercer R., Sylvester R., Wolff J. Temporary remissions in acute leukemia in children produced by folic acid antagonist, 4-aminopteroyl-glutamic acid (aminopterin). *N Engl J Med* 1948;238(23):787–793.

66. Possemato R, Marks KM, Shaul YD, et al. Functional genomics reveal that the serine synthesis pathway is essential in breast cancer. *Nature* 2011;476:346–350.

67. Polyak K, Xia Y, Zweier JL, Kinzler KW, Vogelstein B. A model for p53-induced apoptosis. *Nature* 1997;389:300–305.

68. Liu W, Le A, Hancock C, et al. Reprogramming of proline and glutamine metabolism contributes to the proliferative and metabolic responses regulated by

oncogenic transcription factor c-MYC. *Proc Natl Acad Sci* 2012;109(23):8983–8988.

69. Salimian Rizi B, Achreja A, Nagrath D. Nitric oxide: The forgotten child of tumor metabolism. *Trends in Cancer* 2017;3(9):659–672.

70. Thomas DD, Espey MG, Ridnour LA, et al. Hypoxic inducible factor 1α, extracellular signal-regulated kinase, and p53 are regulated by distinct threshold concentrations of nitric oxide. *Proc Natl Acad Sci* 2004;101(24):8894–8899.

71. Tanese K, Grimm EA, Ekmekcioglu S. The role of melanoma tumor-derived nitric oxide in the tumor inflammatory microenvironment: its impact on the chemokine expression profile , including suppression of CXCL10. *Int J Cancer* 2012;131:891–901.

72. Sanuphan A, Chunhacha P, Pongrakhananon V, Chanvorachote P. Long-term nitric oxide exposure enhances lung cancer cell migration. *Biomed Res Int* 2013;2013:186972.

73. Berchner-Pfannschmidt U, Yamac H, Trinidad B, Fandrey J. Nitric oxide modulates oxygen sensing by hypoxia-inducible factor 1-dependent induction of prolyl hydroxylase 2. *J Biol Chem* 2007;282(3):1788–1796.

74. Nisoli E, Carruba MO. Nitric oxide and mitochondrial biogenesis. *J Cell Sci* 2006;119(14):2855–2862.

75. Russell D, Snyder SH. Amine synthesis in rapidly growing tissues: Ornithine decarboxylase activity in regenerating rat liver, chick embryo, and various tumors. *Proc Natl Acad Sci* 1968;60(4):1420–1427.

76. Gerner EW, Meyskens Jr FL. Polyamines and cancer: Old molecules, new understanding. *Nat Rev Cancer* 2004;4:781–792.

77. García-Faroldi G, Sánchez-Jiménez F, Fajardo I. The polyamine and histamine metabolic interplay in cancer and chronic inflammation. *Curr Opin Clin Nutr Metab Care* 2009;12:59–65.

78. Auvinen M, Paasinen A, Andersson LC, Hölttä E. Ornithine decarboxylase activity is critical for cell transformation. *Nature* 1992;360:355–358.

79. McCann PP, Pegg AE. Ornithine decarboxylase as an enzyme target for therapy. *Pharmacol Ther* 1992;54:195–215.

80. Babbar N, Ignatenko NA, Casero RA, Gerner EW. Cyclooxygenase-independent induction of apoptosis by sulindac sulfone is mediated by polyamines in colon cancer. *J Biol Chem* 2003;278(48):47762–47775.

81. Ignatenko NA, Babbar N, Mehta D, Casero RA, Gerner EW. Suppression of polyamine catabolism by activated Ki-ras in human colon cancer cells. *Mol Carcinog* 2004;39:91–102.

82. Bello-Fernandez C, Packham G, Cleveland JL. The ornithine decarboxylase gene is a transcriptional target of c-Myc. *Proc Natl Acad Sci* 1993;90:7804–7808.

83. Bauer PM, Buga GM, Fukuto JM, Pegg AE, Ignarro LJ. Nitric oxide inhibits ornithine decarboxylase via S-nitrosylation of cysteine 360 in the active site of the enzyme. *J Biol Chem* 2001;276(37):34458–34464.

84. Saulnier Sholler GL, Gerner EW, Bergendahl G, et al. A phase I trial of DFMO targeting polyamine addiction in patients with relapsed/refractory neuroblastoma. *PLoS One* 2015;10(5):e0127246.

85. Zabala-Letona A, Arruabarrena-Aristorena A, Martín-Martín N, et al. mTORC1-dependent AMD1 regulation sustains polyamine metabolism in prostate cancer. *Nature* 2017;547:109–113.

86. Carmeliet P, Jain RK. Angiogenesis in cancer and other diseases. *Nature* 2000;407:249–257.

87. Spolarics Z, Lang CH, Bagby GJ, Spitzer JJ. Glutamine and fatty acid oxidation are the main sources of energy for Kupffer and endothelial cells. *Am J Physiol* 1991;261(2):G185–G190.

88. Leighton B, Curi R, Hussein A, Newsholme EA. Maximum activities of some key enzymes of glycolysis, glutaminolysis, Krebs cycle and fatty acid utilization in bovine pulmonary endothelial cells. *FEBS Lett* 1987;225(1–2):93–96.

89. Dobrina A, Rossi F. Metabolic properties of freshly isolated bovine endothelial cells. Biochim Biophys Acta 1983;762:295–301.

90. Krützfeldt A, Spahr R, Mertens S, Siegmund B, Piper HM. Metabolism of exogenous substrates by coronary endothelial cells in culture. *J Mol Cell Cardiol* 1990;22:1393–1404.

91. Peters K, Kamp G, Berz A, et al. Changes in human endothelial cell energy metabolic capacities during in vitro cultivation. The role of aerobic glycolysis and proliferation. *Cell Physiol Biochem* 2009;24:483–492.

92. Harjes U, Bensaad K, Harris AL. Endothelial cell metabolism and implications for cancer therapy. *Br J Cancer* 2012;107:1207–1212.

93. De Bock K, Georgiadou M, Schoors S, et al. Role of PFKFB3-driven glycolysis in vessel sprouting. *Cell* 2013;154:651–663.

94. Van Hée VF, Pérez-Escuredo J, Cacace A, Copetti T, Sonveaux P. Lactate does not activate NF-κB in oxidative tumor cells. *Front Pharmacol* 2015;6.

95. Leopold JA, Zhang YY, Scribner AW, Stanton RC, Loscalzo J. Glucose-6-phosphate dehydrogenase overexpression decreases endothelial cell oxidant stress and increases bioavailable nitric oxide. *Arterioscler Thromb Vasc Biol* 2003;23(3):411–417.

96. Nacev BA, Grassi P, Dell A, Haslam SM, Liu JO. The antifungal drug itraconazole inhibits Vascular Endothelial Growth Factor Receptor 2 (VEGFR2) glycosylation, trafficking, and signaling in endothelial cells. *J Biol Chem* 2011;286(51):44045–44056.

97. Polet F, Feron O. Endothelial cell metabolism and tumour angiogenesis: glucose and glutamine as essential fuels and lactate as the driving force. *J Intern Med* 2013;273:156–165.

98. Maity P, Chakraborty S, Bhattacharya P. Angiogenesis - A putative new approach in glutamine related therapy. *Pathol Oncol Res* 1999;5(4):309–314.

99. Huang H, Vandekeere S, Kalucka J, et al. Role of glutamine and interlinked asparagine metabolism in vessel formation. *EMBO J* 2017;1–19.

100. Kim B, Li J, Jang C, Arany Z. Glutamine fuels proliferation but not migration of endothelial cells. *EMBO J* 2017;36:2321–2333.

101. Morrison RF, Seidel ER. Vascular endothelial cell proliferation: regulation of cellular polyamines. *Cardiovasc Res* 1995;29:841–847.

102. Urdiales JL, Medina MA, Sánchez-Jiménez F. Polyamine metabolism revisited. *Eur J Gastroenterol Hepatol 2001*;13(9):1015–1019.

103. Li H, Meininger CJ, Bazer FW, Wu G. Intracellular sources of ornithine for polyamine synthesis in endothelial cells. *Amino Acids* 2016;48:2401–2410.

104. Wu G, Haynes TE, Li H, Meininger CJ. Glutamine metabolism in endothelial cells: ornithine synthesis from glutamine via pyrroline-5-carboxylate synthase. *Comp Biochem Physiol Part A Mol Integr Physiol* 2000;126(1):115–123.

105. Spahr R, Krtitzfeldt A, Mertens S, Siegmund B, Piper HM. Fatty acids are not an important fuel for coronary microvascular endothelial cells. *Mol Cell Biochem* 1989;88:59–64.

106. Schoors S, Bruning U, Missiaen R, Queiroz KCS. Fatty acid carbon is essential for dNTP synthesis in endothelial cells. *Nature* 2015;520(7546):192–197.

107. Missiaen R, Rodriguez FM, Eelen G, Carmeliet P. Targeting endothelial

metabolism for anti-angiogenesis therapy: A pharmacological perspective. *Vascul Pharmacol* 2017;90:8–18.

108. De Bock K, Georgiadou M, Carmeliet P. Role of endothelial cell metabolism in vessel sprouting. *Cell Metab* 2013;18(5):634–647.

109. Eelen G, Cruys B, Welti J, De Bock K, Carmeliet P. Control of vessel sprouting by genetic and metabolic determinants. *Trends Endocrinol Metab* 2013;24(12):589–596.

110. Potente M, Carmeliet P. The link between angiogenesis and endothelial metabolism. *Annu Rev Physiol* 2017;79:43–66.

111. Cadamuro M, Nardo G, Indraccolo S, et al. Platelet-derived growth factor-D and Rho GTPases regulate recruitment of cancer-associated fibroblasts in cholangiocarcinoma. *Hepatology* 2013;58(3):1042–1053.

112. Wagner EF. Cancer: Fibroblasts for all seasons. *Nature* 2016;530(7588):42–43.

113. Koliaraki V, Pasparakis M, Kollias G. IKKβ in intestinal mesenchymal cells promotes initiation of colitis-associated cancer. *J Exp Med* 2015;212(13):2235–2251.

114. Pallangyo CK, Ziegler PK, Greten FR. IKKβ acts as a tumor suppressor in cancer-associated fibroblasts during intestinal tumorigenesis. *J Exp Med* 2015;212(13):2253–2266.

115. Carito V, Bonuccelli G, Martinez-Outschoorn UE, et al. Metabolic remodeling of the tumor microenvironment: Migration stimulating factor (MSF) reprograms myofibroblasts toward lactate production, fueling anabolic tumor growth. *Cell Cycle* 2012;11(18):3403–3414.

116. Whitaker-Menezes D, Martinez-Outschoorn UE, Lin Z, et al. Evidence for a stromal-epithelial "lactate shuttle" in human tumors: MCT4 is a marker of oxidative stress in cancer-associated fibroblasts. *Cell Cycle* 2011;10(11):1772–1783.

117. Zhang D, Wang Y, Shi Z, et al. Metabolic reprogramming of cancer-associated fibroblasts by IDH3α downregulation. *Cell Rep* 2015;10:1335–1348.

118. Koukourakis MI, Giatromanolaki A, Harris AL, Sivridis E. Comparison of metabolic pathways between cancer cells and stromal cells in colorectal carcinomas: A metabolic survival role for tumor-associated stroma. *Cancer Res* 2006;66(2):632–637.

119. Rattigan YI, Patel BB, Ackerstaff E, et al. Lactate is a mediator of metabolic cooperation between stromal carcinoma associated fibroblasts and glycolytic

tumor cells in the tumor microenvironment. *Exp Cell Res* 2012;318:326–335.

120. Yang L, Achreja A, Yeung TL, et al. Targeting stromal glutamine synthetase in tumors disrupts tumor microenvironment-regulated cancer cell growth. *Cell Metab* 2016;24:685–700.

121. Liu Y, Cao X. The origin and function of tumor-associated macrophages. *Cell Mol Immunol* 2015;12:1–4.

122. Quatromoni JG, Eruslanov E. Tumor-associated macrophages: function, phenotype, and link to prognosis in human lung cancer. *Am J Transl Res* 2012;4(4):376–389.

123. Grivennikov SI, Greten FR, Karin M. Immunity, inflammation, and cancer. *Cell* 2010;140(6):883–899.

124. Wang YC, He F, Feng F, et al. Notch signaling determines the M1 versus M2 polarization of macrophages in antitumor immune responses. *Cancer Res* 2010;70(12):4840–4849.

125. Franklin RA, Liao W, Sarkar A, et al. The cellular and molecular origin of tumor-associated macrophages. *Science* 2014;344(6186):921–925.

126. Chittezhath M, Dhillon MK, Lim JY, et al. Molecular profiling reveals a tumor-promoting phenotype of monocytes and macrophages in human cancer progression. *Immunity* 2014;41:815–829.

127. Lampropoulou V, Sergushichev A, Bambouskova M, et al. Itaconate Links Inhibition of Succinate Dehydrogenase with Macrophage Metabolic Remodeling and Regulation of Inflammation. *Cell Metab* 2016;24:158–166.

128. Rodríguez-Prados J-C, Través PG, Cuenca J, et al. Substrate fate in activated macrophages: a comparison between innate, classic, and alternative activation. *J Immunol* 2010;185:605–614.

129. Tannahill GM, Curtis AM, Adamik J, et al. Succinate is an inflammatory signal that induces IL-1β through HIF-1α. *Nature* 2013;496(7444):238–242.

130. Selak MA, Armour SM, MacKenzie ED, et al. Succinate links TCA cycle dysfunction to oncogenesis by inhibiting HIF-α prolyl hydroxylase. *Cancer Cell* 2005;7(1):77–85.

131. Luo W, Hu H, Chang R, et al. Pyruvate kinase M2 is a PHD3-stimulated coactivator for hypoxia-inducible factor 1. *Cell* 2011;145:732–744.

132. Alves-Filho JC, Pålsson-McDermott EM. Pyruvate Kinase M2: A Potential Target for Regulating Inflammation. *Front Immunol* 2016;7:1–7.

133. Kelly B, O'Neill LA. Metabolic reprogramming in macrophages and dendritic cells in innate immunity. *Cell Res* 2015;25(7):771–784.

134. Ocaña MC, Martínez-Poveda B, Quesada AR, Medina MA. Metabolism in the tumor microenvironment: What is known about stromal and immune cells? *Clin Immunol Endocr Metab Drugs* 2017;4.

135. Arts RJW, Plantinga TS, Tuit S, et al. Transcriptional and metabolic reprogramming induce an inflammatory phenotype in non-medullary thyroid carcinoma-induced macrophages. *Oncoimmunology* 2016;5(12):e1229725.

136. Liu D, Chang C, Lu N, et al. Comprehensive proteomics analysis reveals metabolic reprogramming of tumor-associated macrophages stimulated by the tumor microenvironment. *J Proteome Res* 2017;16:288–297.

137. Daurkin I, Eruslanov E, Stoffs T, et al. Tumor-associated macrophages mediate immunosuppression in the renal cancer microenvironment by activating the 15-lipoxygenase-2 pathway. *Microenviron Immunol* 2011;71(20):6400–6410.

138. Choi J, Stradmann-bellinghausen B, Yakubov E, Savaskan NE, Anne R. Glioblastoma cells induce differential glutamatergic gene expressions in human tumor-associated microglia/macrophages and monocyte-derived macrophages. *Cancer Biol Ther* 2015;16(8):1205–1213.

139. Covarrubias AJ, Aksoylar HI, Yu J, et al. Akt-mTORC1 signaling regulates Acly to integrate metabolic input to control of macrophage activation. *eLife* 2016;5:e11612.

140. Covarrubias AJ, Aksoylar HI, Horng T. Control of macrophage metabolism and activation by mTOR and Akt signaling. *Semin Immunol* 2015;27(4):286–296.

141. Delgoffe GM, Powell JD. Sugar, fat, and protein: New insights into what T cells crave. *Curr Opin Immunol* 2015;33:49–54.

142. Pennisi E. Metabolic shift may train immune cells. *Science* 2014;345(6204):1550–1551.

143. Kouidhi S, Elgaaied AB, Chouaib S. Impact of Metabolism in on T-Cell Differentiation and Function and Cross Talk with Tumor Microenvironment. *Front Immunol* 2017;8:270.

144. Hubler MJ, Kennedy AJ. Role of Lipids in the Metabolism and Activation of Immune Cells. *J Nutr Biochem* 2016;34:1–7.

145. Ho PC, Liu PS. Metabolic communication in tumors: a new layer of immunoregulation for immune evasion. *J Immunother Cancer* 2016;4(1):1.

146. Ho PC, Bihuniak JD, MacIntyre AN, et al. Phosphoenolpyruvate is a metabolic checkpoint of anti-tumor T cell responses. *Cell* 2015;162:1–12.

147. Ganeshan K, Chawla A. Metabolic regulation of immune responses. *Annu Rev Immunol* 2014;32(1):609–634.

148. Delgoffe GM, Kole TP, Zheng Y, et al. The mTOR kinase differentially regulates effector and regulatory T cell lineage commitment. *Immunity* 2009;30:832–844.

149. Dang E V., Barbi J, Yang HY, et al. Control of TH17/Treg balance by hypoxia-inducible factor 1. *Cell* 2011;146:772–784.

150. Wang R, Dillon CP, Shi LZ, et al. The transcription factor Myc controls metabolic reprogramming upon T lymphocyte activation. *Immunity* 2011;35:871–882.

151. Newsholme P. Why is L-glutamine metabolism important to cells of the immune system in health , postinjury , surgery or infection? *J Nutr* 2001;131:2515S–2522S.

152. Geiger R, Rieckmann JC, Wolf T, et al. L-Arginine modulates T cell metabolism and enhances survival and anti-tumor activity. *Cell* 2016;167:829–842.

153. Varricchi G, Galdiero MR, Loffredo S, et al. Are mast cells MASTers in cancer? *Front Immunol* 2017;8:424.

154. Liu J, Zhang Y, Zhao J, et al. Mast cell: insight into remodeling a tumor microenvironment. *Cancer Metastasis Rev* 2011;30:177–184.

155. Ribatti D, Crivellato E. Mast cells, angiogenesis, and tumour growth. *Biochim Biophys Acta* 2012;1822:2–8.

156. Norrby K, Jakobsson A, Sörbo J. Mast-cell secretion and angiogenesis, a quantitative study in rats and mice. *Virchows Arch B Cell Pathol* 1989;57:251–256.

157. Chakravarty N. Glycolysis in rat peritoneal mast cells. *J Cell Biol* 1965;25:123–128.

158. Chakravarty N. Further observations on the inhibition of histamine release by 2-deoxyglucose. *Acta Physiol Scand* 1968;72:425–432.

159. Chakravarty N, Sorensen J. Stimulation of glucose metabolism in rat mast cells by antigen, dextran and compound 48/80, used as histamine releasing agents. *Acta Physiol Scand* 1974;91:339–353.

160. Yoshizaki K, Arizono N, Hayano T, Watari H. Allergen-induced histamine

secretion associated with lactate production in mast cells detected by 1H-NMR. *Magn Reson Med* 1993;29:732–736.

161. Johansen T. Dependence of anaphylactic histamine release from rat mast cells on cellular energy metabolism. *Eur J Pharmacol* 1981;72:281–286.

162. Mitra R, Pal S. Inhibition of mast cell population by L-glutamine in aspirin-induced ulceration in rat stomach. *Indian J Physiol Pharmacol* 1977;21(4):374–378.

163. Lechowski S, Feilhauer K, Staib L, Coëffier M, Bischoff SC, Lorentz A. Combined arginine and glutamine decrease release of de novo synthesized leukotrienes and expression of proinflammatory cytokines in activated human intestinal mast cells. *Eur J Nutr* 2013;52:505–512.

164. Kawasaki H, Chang H., Tseng H., et al. A tryptophan metabolite , kynurenine, promotes mast cell activation through aryl hydrocarbon receptor. *Allergy* 2014;69:445–452.

165. Opitz CA, Litzenburger UM, Sahm F, et al. An endogenous tumour-promoting ligand of the human aryl hydrocarbon receptor. *Nature* 2011;478:197–203.

166. Sekar Y, Moon TC, Slupsky CM, Befus AD. Protein tyrosine nitration of aldolase in mast cells: A plausible pathway in nitric oxide-mediated regulation of mast cell function. *J Immunol* 2010;185:578–587.

167. Ryu SY, Hong GU, Kim DY, Ro JY. Enolase 1 and calreticulin regulate the differentiation and function of mouse mast cells. Cell Signal 2012;24:60–70.

168. Sharkia I, Erlich TH, Landolina N, et al. Pyruvate dehydrogenase has a major role in mast cell function, and its activity is regulated by mitochondrial microphthalmia transcription factor. *J Allergy Clin Immunol* 2017;140(1):204–214.

169. Zheng M, Cho D-I, Le HT, Cheon SH, Kim K-M. Dual regulation of mast cell degranulation through IgE receptor-mediated modulation of M2-type pyruvate kinase. *Arch Pharm Res* 2014;37:1177–1182.

170. García-Faroldi G, Rodríguez CE, Urdiales JL, et al. Polyamines are present in mast cell secretory granules and are important for granule homeostasis. *PLoS One* 2010;5(11):e15071.

171. Hosono J, Morikawa S, Ezaki T, Kawamata T, Okada Y. Pericytes promote abnormal tumor angiogenesis in a rat RG2 glioma model. *Brain Tumor Pathol* 2017;34(3):120–129.

172. Caspani EM, Crossley PH, Redondo-Garcia C, Martinez S. Glioblastoma: A

pathogenic crosstalk between tumor Cells and pericytes. *PLoS One* 2014;9(7):e101402.

173. Ribeiro AL, Kaid C, Silva PBG, Cortez BA, Okamoto OK. Inhibition of lysyl oxidases impairs migration and angiogenic properties of tumor-associated pericytes. *Stem Cells Int* 2017;2017.

174. Berrone E, Beltramo E, Solimine C, Ape AU, Porta M. Regulation of intracellular glucose and polyol pathway by thiamine and benfotiamine in vascular cells cultured in high glucose. *J Biol Chem* 2006;281(14):9307–9313.

175. Trudeau K, Molina AJA, Roy S. High glucose induces mitochondrial morphology and metabolic changes in retinal pericytes. *Invest Ophthalmol Vis Sci* 2011;52(12):8657–8664.

176. Yuan K, Shao N, Hennigs JK, et al. Increased pyruvate dehydrogenase kinase 4 expression in lung pericytes is associated with reduced endothelial-pericyte interactions and small vessel loss in pulmonary arterial hypertension. *Am J Pathol* 2016;186(9):2500–2514.

177. Hanahan D, Coussens LM. Accessories to the crime: Functions of cells recruited to the tumor microenvironment. *Cancer Cell* 2012;21:309–322.

178. Burnet FM. The concept of immunological surveillance. *Prog Exp Tumor Res* 1970;13:1–27.

179. Thomas L. On immunosurveillance in human cancer. *Yale J Biol Med* 1982;55:329–333.

180. Sukumar M, Roychoudhuri R, Restifo NP. Nutrient competition: A new axis of tumor immunosuppression. *Cell* 2015;162:1206–1208.

181. Chang C, Curtis JD, Maggi Jr LB, et al. Posttranscriptional control of T cell effector function by aerobic glycolysis. *Cell* 2013;153:1239-1251.

182. Fischer K, Hoffmann P, Voelkl S, et al. Inhibitory effect of tumor cell–derived lactic acid on human T cells. *Blood* 2007;109(9):3812–3820.

183. Chaudhary B, Elkord E. Regulatory T cells in the tumor microenvironment and cancer progression: Role and therapeutic targeting. *Vaccines* 2016;4:28.

184. Ahmadzadeh M, Johnson LA, Heemskerk B, et al. Tumor antigen − specific CD8 T cells infiltrating the tumor express high levels of PD-1 and are functionally impaired Tumor antigen – specific CD8 T cells infiltrating the tumor express high levels of PD-1 and are functionally impaired. *Blood* 2009;114:1537–1544.

185. Dong H, Strome SE, Salomao DR, et al. Tumor-associated B7-H1 promotes T-

cell apoptosis: A potential mechanism of immune evasion. *Nat Med* 2002;8(8):793–800.

186. Parry RV, Chemnitz JM, Frauwirth KA, et al. CTLA-4 and PD-1 Receptors Inhibit T-Cell Activation by Distinct Mechanisms CTLA-4 and PD-1 Receptors Inhibit T-Cell Activation by Distinct Mechanisms. *Mol Cell Biol* 2005;25(21):9543–9553.

187. Patsoukis N, Bardhan K, Chatterjee P, et al. PD-1 alters T-cell metabolic reprogramming by inhibiting glycolysis and promoting lipolysis and fatty acid oxidation. *Nat Commun* 2015;6:6692.

188. Fallarino F, Grohmann U, You S, et al. The combined effects of tryptophan starvation and tryptophan catabolites down-regulate T cell receptor ζ-chain and induce a regulatory phenotype in naive T cells. *J Immmunology* 2006;176:6752–6761.

189. Häusler SFM, Montalbán Del Barrio I, Strohschein J, et al. Ectonucleotidases CD39 and CD73 on OvCA cells are potent adenosine-generating enzymes responsible for adenosine receptor 2A-dependent suppression of T cell function and NK cell cytotoxicity. *Cancer Immunol Immunother* 2011;60:1405–1418.

190. Salimian Rizi B, Caneba C, Nowicka A, et al. Nitric oxide mediates metabolic coupling of omentum-derived adipose stroma to ovarian and endometrial cancer cells. *Cancer Res* 2015;75:456–471.

191. Folkman J. Tumor angiogenesis: therapeutic implications. *N Engl J Med* 1971;285(21):1182–1186.

192. Folkman J. Angiogenesis: an organizing principle for drug discovery? *Nat Rev Drug Discov* 2007;6:273–286.

193. Ribatti D, Nico B, Crivellato E, Roccaro AM, Vacca A. The history of the angiogenic switch concept. *Leukemia* 2007;21:44–52.

194. Chappell JC, Wiley DM, Bautch VL. Regulation of blood vessel sprouting. *Semin Cell Dev Biol* 2011;22:1005–1011.

195. Carmeliet P. Angiogenesis in life, disease and medicine. *Nature* 2005;438:932–936.

196. Potente M, Gerhardt H, Carmeliet P. Basic and therapeutic aspects of angiogenesis. *Cell* 2011;146:873–87.

197. Goveia J, Stapor P, Carmeliet P. Principles of targeting endothelial cell metabolism to treat angiogenesis and endothelial cell dysfunction in disease. *EMBO Mol Med* 2014;6(9):1105–1120.

198. Végran F, Boidot R, Michiels C, Sonveaux P, Feron O. Lactate influx through the endothelial cell monocarboxylate transporter MCT1 supports an NF-kB/IL-8 pathway that drives tumor angiogenesis. *Cancer Res* 2011;71(7):2550–2560.

199. Sonveaux P, Copetti T, de Saedeleer CJ, et al. Targeting the lactate transporter MCT1 in endothelial cells inhibits lactate-induced HIF-1 activation and tumor angiogenesis. *PLoS One* 2012;7(3):e33418.

200. Ruan GX, Kazlauskas A. Lactate engages receptor tyrosine kinases Axl, Tie2, and vascular endothelial growth factor receptor 2 to activate phosphoinositide 3-kinase/AKT and promote angiogenesis. *J Biol Chem* 2013;288(29):21161–21172.

201. Reihill JA, Ewart M-A, Salt IP. The role of AMP-activated protein kinase in the functional effects of vascular endothelial growth factor-A and -B in human aortic endothelial cells. *Vasc Cell* 2011;3(1):9.

202. Yamanishi S, Katsumura K, Kobayashi T, Puro DG. Extracellular lactate as a dynamic vasoactive signal in the rat retinal microvasculature. *Am J Physiol Hear Circ Physiol* 2006;290:H925–H934.

203. Hayakawa Y, Wang TC. Nerves switch on angiogenic metabolism. *Science* 2017;358(6361):305–306.

204. Zahalka AH, Arnal-Estapé A, Maryanovich M, et al. Adrenergic nerves activate an angio-metabolic switch in prostate cancer. *Science* 2017;358(6361):321–326.

205. Martinez-Outschoorn UE, Pestell RG, Howell A, et al. Energy transfer in "parasitic" cancer metabolism. *Cell Cycle* 2011;10(24):4208–4216.

206. Pavlides S, Whitaker-Menezes D, Castello-Cros R, et al. The reverse Warburg effect: Aerobic glycolysis in cancer associated fibroblasts and the tumor stroma. *Cell Cycle* 2009;8(23):3984–4001.

207. Bonuccelli G, Avnet S, Grisendi G, et al. Role of mesenchymal stem cells in osteosarcoma and metabolic reprogramming of tumor cells. *Oncotarget* 2014;5(17):7575–7588.

208. Romero-García S, Moreno-Altamirano MMB, Prado-García H, Sánchez-García FJ. Lactate contribution to the tumor microenvironment: Mechanisms, effects on immune cells and therapeutic relevance. *Front Immunol* 2016;7.

209. Stern R, Shuster S, Neudecker BA, Formby B. Lactate stimulates fibroblast expression of hyaluronan and CD44: the Warburg effect revisited. *Exp Cell Res* 2002;276:24–31.

210. Scherz-Shouval R, Santagata S, Mendillo ML, et al. The reprogramming of tumor stroma by HSF1 is a potent enabler of malignancy. *Cell* 2014;158:564–

578.

211. Orimo A, Gupta PB, Sgroi DC, et al. Stromal fibroblasts present in invasive human breast carcinomas promote tumor growth and angiogenesis through elevated SDF-1/CXCL12 secretion. *Cell* 2005;121:335–348.

212. Attieh Y, Vignjevic DM. The hallmarks of CAFs in cancer invasion. *Eur J Cell Biol* 2016;95:493–502.

213. Roy A, Bera S. CAF cellular glycolysis: linking cancer cells with the microenvironment. *Tumor Biol* 2016;37:8503–8514.

214. Guo X, Oshima H, Kitmura T, Taketo MM, Oshima M. Stromal fibroblasts activated by tumor cells promote angiogenesis in mouse gastric cancer. *J Biol Chem* 2008;283(28):19864–19871.

215. Zhang Y, Tang H, Cai J, et al. Ovarian cancer-associated fibroblasts contribute to epithelial ovarian carcinoma metastasis by promoting angiogenesis, lymphangiogenesis and tumor cell invasion. *Cancer Lett* 2011;303:47–55.

216. Lopes-Coelho F, André S, Félix A, Serpa J. Breast cancer metabolic cross-talk: Fibroblasts are hubs and breast cancer cells are gatherers of lipids. *Mol Cell Endocrinol* 2017.

217. Augsten M, Sjöberg E, Frings O, et al. Cancer-associated fibroblasts expressing CXCL14 rely upon NOS1-derived nitric oxide signaling for their tumor-supporting properties. *Cancer Res* 2014;74:2999–3010.

218. Penny HL, Sieow JL, Adriani G, et al. Warburg metabolism in tumor-conditioned macrophages promotes metastasis in human pancreatic ductal adenocarcinoma. *Oncoimmunology* 2016;5(8):e1191731.

219. Colegio OR, Chu N-Q, Szabo AL, et al. Functional polarization of tumour-associated macrophages by tumour-derived lactic acid. *Nature* 2014;513(7519):559–563.

220. Leek RD, Lewis CE, Whitehouse R, Greenall M, Clarke J, Harris AL. Association of macrophage infiltration with angiogenesis and prognosis in invasive breast carcinoma. *Cancer Res* 1996;56:4625–4629.

221. Pollard JW. Tumour-educated macrophages promote tumour progression and metastasis. *Nat Rev Cancer* 2004;4:71–78.

222. Lin L, Chen YS, Yao YD, et al. CCL18 from tumor-associated macrophages promotes angiogenesis in breast cancer. *Oncotarget* 2015;6(33):34758–34773.

223. Casazza A, Laoui D, Wenes M, et al. Impeding macrophage entry into hypoxic

tumor areas by Sema3A/Nrp1 signaling blockade inhibits angiogenesis and restores antitumor immunity. *Cancer Cell* 2013;24:695–709.

224. Yuan Y, Jiang YC, Sun CK, Chen QM. Role of the tumor microenvironment in tumor progression and the clinical applications. *Oncol Rep* 2016;35:2499–2515.

225. Rabold K, Netea MG, Adema GJ, Netea-Maier RT. Cellular metabolism of tumor-associated macrophages: functional impact and consequences. *FEBS Lett* 2017.

226. Albina E. Modulation by products of glucose metabolism in macrophages of nitric oxide synthase. *Am J Physiol* 1993;264:C1594–C1599.

227. Klimp AH, Hollema H, Kempinga C, van der Zee AGJ, de Vries EGE, Daemen T. Expression of cyclooxygenase-2 and inducible nitric oxide synthase in human ovarian tumors and tumor-associated macrophages. *Cancer Res* 2001;61:7305–7309.

228. DiNapoli MR, Calderón CL, López DM. The altered tumoricidal capacity of macrophages isolated from tumor-bearing mice is related to reduced expression of the inducible nitric oxide synthase gene. *J Exp Med* 1996;183:1323–1329.

229. Kannan Y, Perez-Lloret J, Li Y, et al. TPL-2 regulates macrophage lipid metabolism and M2 differentiation to control TH2-mediated immunopathology. *PLoS Pathog* 2016;12(8):e1005783.

230. Zhao Q, Kuang D, Wu Y, et al. Activated CD69+ T cells foster immune privilege by regulating IDO expression in tumor-associated macrophages. *J Immunol* 2011;188:1117–1124.

231. Zhu Q, Wu X, Wu Y, Wang X. Interaction between Treg cells and tumor-associated macrophages in the tumor microenvironment of epithelial ovarian cancer. *Oncol Rep* 2016;36:3472–3478.

232. Kabat AM, Pearce EJ. Inflammation by way of macrophage metabolism. *Science* 2017;356(6337):488–489

233. Ip WKE, Hoshi N, Shouval DS, Snapper S. Anti-inflammatory effect of IL-10 mediated by metabolic reprogramming of macrophages. *Science* 2017;356:513–519.

234. Noman MZ, Desantis G, Janji B, et al. PD-L1 is a novel direct target of HIF-1, and its blockade under hypoxia enhanced MDSC-mediated T cell activation. *J Exp Med* 2014;211(5):781–790.

235. Shime H, Yabu M, Akazawa T, et al. Tumor-Secreted Lactic Acid Promotes IL-23/IL-17 Proinflammatory Pathway. *J Immunol* 2008;180:7175–7183.

236. Pasquier J, Guerrouahen BS, Al Thawadi H, et al. Preferential transfer of mitochondria from endothelial to cancer cells through tunneling nanotubes modulates chemoresistance. *J Transl Med* 2013;11(1):94.

237. Kamphorst JJ, Gottlieb E. Cancer metabolism: Friendly neighbours feed tumour cells. *Nature* 2016;536:401–402.

238. Sousa CM, Biancur DE, Wang X, et al. Pancreatic stellate cells support tumour metabolism through autophagic alanine secretion. *Nature* 2016;536:479–83.

239. Zhang W, Trachootham D, Liu J, et al. Stromal control of cystine metabolism promotes cancer cell survival in chronic lymphocytic leukaemia. *Nat Cell Biol* 2012;14(3):276–286.

240. Meyer KA, Neeley CK, Baker NA, et al. Adipocytes promote pancreatic cancer cell proliferation via glutamine transfer. *Biochem Biophys Reports* 2016;7:144–149.

241. Gazi E, Gardner P, Lockyer NP, Hart CA, Brown MD, Clarke NW. Direct evidence of lipid translocation between adipocytes and prostate cancer cells with imaging FTIR microspectroscopy. *J Lipid Res* 2007;48:1846–1856.

242. Nieman KM, Kenny HA, Penicka CV, et al. Adipocytes promote ovarian cancer metastasis and provide energy for rapid tumor growth. *Nat Med* 2011;17(11):1498–1503.

243. Wen Y-A, Xing X, Harris JW, et al. Adipocytes activate mitochondrial fatty acid oxidation and autophagy to promote tumor growth in colon cancer. *Cell Death Dis* 2017;8(2):e2593.

244. Dirat B, Bochet L, Dabek M, et al. Cancer-associated adipocytes exhibit an activated phenotype and contribute to breast cancer invasion. *Cancer Res* 2011;71(7):2455–2465.

245. Choi MS, Jung J, Kim H, Ham MR, Lee TR, Shin DW. S-nitrosylation of fatty acid synthase regulates its activity through dimerization. *J Lipid Res* 2016;57:607–615.

246. Al-zhoughbi W, Huang J, Paramasivan GS, Till H, Pichler M, Guertl-lackner B. Tumor macroenvironment and metabolism. *Semin Oncol* 2014;41(2):281–295.

247. Shapot VS. Systemic effect of the tumor on the host: Biochemical and endocrine manifestations. *Adv Enzyme Regul* 1975;13:67–75.

248. Lee Y, Chang W-C, Ma W-L. Hypothesis: solid tumours behave as systemic metabolic dictators. *J Cell Mol Med* 2016;20(6):1076–1085.

249. Shapot VS. Biochemical aspects of tumour growth. Mir Publishers; 1980.

250. Mider GB. Some aspects of nitrogen and energy metabolism in cancerous subjects: A review. *Cancer Res* 1951;11:821–829.

251. Souba WW. Glutamine and Cancer. *Ann Surg* 1993;218(6):715–728.

252. Wu C, Morris HP. Responsiveness of glutamine-metabolizing enzymes in Morris hepatomas to metabolic modulations. *Cancer Res* 1970;30:2675–2684.

253. Collins CL, Wasa M, Souba WW, Abcouwer SF. Regulation of glutamine synthetase in human breast carcinoma cells and experimental tumors. *Surgery* 1997;122(2):451–463.

254. Labow BI, Souba WW, Abcouwer SF. Mechanisms governing the expression of the enzymes of glutamine metabolism - glutaminase and glutamine synthetase. *J Nutr* 2001;131:2467S–2474S.

255. Márquez J, Sánchez-Jiménez F, Medina MA, Quesada AR, Núñez de Castro I. Nitrogen metabolism in tumor bearing mice. *Arch Biochem Biophys* 1989;268(2):667–675.

256. Vornovitskaya GI, Dubinina IG, Gershtein ES, Grekhova N V, Shapot VS. Changes in relations between two pathways of synthesis of RNA precursors in the tissues of animals with fast growing hepatomas. *Bull Exp Biol Med* 1979;87(3):264–266.

257. Gershtein ES, Vornovitskaya GI, Shapot VS. Kinetics of (C14) thymidine metabolism in hepatomas and tissues from normal and tumor-bearing animals. *Biokhimiia* 1978;43(7):1303–1311.

258. Medina MA. Glutamine and Cancer. *J Nutr* 2001;131(9):2539S–2542S.

259. Yoshida S, Kaibara A, Yamasaki K, Ishibashi N, Noake T, Kakegawa T. Effect of glutamine supplementation on protein metabolism and glutathione in tumor-bearing rats. *J Parenter Enter Nutr* 1995;19:492–497.

260. Ziegler TR. Glutamine supplementation in cancer patients receiving bone marrow transplantation and high dose chemotherapy. *J Nutr* 2001;131(9):2578S–2584S.

261. LePage GA, Potter VR, Busch H, Heidelberger C, Hurlbert RB. Growth of carcinoma implants in fed and fasted rats. *Cancer Res* 1952;12:153–157.

262. Torosian MH, Nguyen HQ. Tumors - Effective nitrogen traps independent of protein Intake. *J Surg Res* 1989;47:456–459.

263. De Lerma Barbaro A. The complex liaison between cachexia and tumor burden.

Oncol Rep 2015;34:1635–1649.

264. Saez S. Adrenal function in cancer: Relation to the evolution. *Eur J Cancer* 1971;7:381–387.

265. Grieninger G, Hertzberg KM, Pindyck J. Fibrinogen synthesis in serum-free hepatocyte cultures: Stimulation by glucocorticoids. *Proc Natl Acad Sci* 1978;75(11):5506–5510.

266. Ioannesyants IA, Adler V V, Él'kina ZI, Artamonova SI, Kadagidze ZG, Shapot VS. Sensitivity of RNA-synthesizing system of the lymphocytes of patients with malignant neoplasms to phytohemagglutinin and dexamethasone. *Bull Exp Biol Med* 1977;83(4):529–532.

267. Flint TR, Janowitz T, Connell CM, et al. Tumor-induced IL-6 reprograms host metabolism to suppress anti-tumor immunity. *Cell Metab* 2016;24:672–684.

268. Cahlin C, Körner A, Axelsson H, Wang W, Lundholm K, Svanberg E. Experimental cancer cachexia: The role of host-derived cytokines interleukin (IL)-6, IL-12, interferon-γ, and tumor necrosis factor α evaluated in gene knockout, tumor-bearing mice on C57 Bl background and eicosanoid-dependent cachexia. *Cancer Res* 2000;60:5488–5493.

269. Masri S, Papagiannakopoulos T, Kinouchi K, et al. Lung adenocarcinoma distally rewires hepatic circadian homeostasis. *Cell* 2016;165:896–909.

270. Shapot VS, Blinov VA. Blood glucose levels and gluconeogenesis in animals bearing transplantable tumors. *Cancer Res* 1974;34:1827–1832.

271. Stumvoll M, Meyer C, Perriello G, Kreider M, Welle S, Gerich J. Human kidney and liver gluconeogenesis: evidence for organ substrate selectivity. *Am J Physiol* 1998;274:E817–E826.

272. Nurjhan N, Bucci A, Perriello G, et al. Glutamine: A major gluconeogenic precursor and vehicle for interorgan carbon transport in man. *J Clin Invest* 1995;95:272–277.

273. Richard V, Conotte R, Mayne D, Colet J. Does the 1H-NMR plasma metabolome reflect the host-tumor interactions in human breast cancer? *Oncotarget* 2017;8(30):49915–49930.

274. Argilés JM, Busquets S, Stemmler B, López-soriano FJ. Cancer cachexia: understanding the molecular basis. *Nat Rev Cancer* 2014;14:754–762.

275. Clark CM, Goodlad GAJ. Muscle protein biosynthesis in the tumour-bearing rat. A defect in a post-initiation stage of translation. *Biochim Biophys Acta* 1975;378:230–240.

276. Pushkina IP, Krechetova CD, Shapot VS. Correlation of membrane-bound and free ribosomes in normal rat liver, Zajdela hepatoma rat liver and ascite cells proper. *Biokhimiia* 1976;41:1940–1944.

277. Baker N, Hill V, Ookhtens M. Regulation of plasma-free fatty acid mobilization by dietary glucose in Ehrlich ascites tumor-bearing mice. *Cancer Res* 1978;38:2372–2377.

278. Das SK. Adipose triglyceride lipase contributes to cancer-associated cachexia. *Science* 2011;333:233–238.

279. Liu L, Wang Y, Zheng T, Cao B, Li M, Shi J, et al. Metabolic shifts induced by human H460 cells in tumor-bearing mice. *Biomed Chromatogr* 2016;30:337–342.

280. Oka T, Ohwada K, Nagao M, Kitazato K. Effect of arginine-enriched total parenteral nutrition host-tumor interaction in cancer-bearing rats. *J Parenter Enter Nutr* 1993;17:375–383.

281. Tachibana K, Mukai K, Hiraoka I, Moriguchi S, Takama S, Kishino Y. Evaluation of the effect of arginine-enriched amino acid solution on tumor growth. *J Parenter Enter Nutr* 1985;9:425–434.

282. Márquez J, Matés JM, Quesada AR, Medina MA, Núñez de Castro I, Sánchez-Jiménez F. Altered ornithine metabolism in tumor-bearing mice. *Life Sci* 1989;45:1877–1884.

283. Ohmura M, Hishiki T, Yamamoto T, et al. Impacts of CD44 knockdown in cancer cells on tumor and host metabolic systems revealed by quantitative imaging mass spectrometry. *Nitric Oxide* 2014;46:102–113.

284. Commisso C, Davidson SM, Soydaner-Azeloglu RG, et al. Macropinocytosis of protein is an amino acid supply route in Ras-transformed cells. *Nature* 2013;497(7451):633–637.

285. Kamphorst JJ, Nofal M, Commisso C, et al. Human pancreatic cancer tumors are nutrient poor and tumor cells actively scavenge extracellular protein. *Cancer Res* 2015;75(3):544–554.

286. Holm E, Hagmãller E, Staedt U, et al. Substrate balances across colonic carcinomas in humans. *Cancer Res* 1995;55:1373–1378.

287. Krishna S, Palm W, Lee Y, et al. PIKfyve regulates vacuole maturation and nutrient recovery following engulfment. *Dev Cell* 2016;38(5):536–547.

288. Palm W, Park Y, Wright K, Pavlova NN, Tuveson, David A, Thompson CB. The utilization of extracellular proteins as nutrients is suppressed by mTORC1. *Cell*

2015;162:259–270.

289. Kamphorst JJ, Cross JR, Fan J, et al. Hypoxic and Ras-transformed cells support growth by scavenging unsaturated fatty acids from lysophospholipids. *Proc Natl Acad Sci* 2013;110(22):8882–8887.

290. Desai N, Trieu V, Yao Z, et al. Increased antitumor activity, intratumor paclitaxel concentrations, and endothelial cell transport of cremophor-free, albumin-bound paclitaxel, ABI-007, compared with cremophor-based paclitaxel. *Clin Cancer Res* 2006;12(4):1317–1325.

291. Von Hoff DD, Ramanathan RK, Borad MJ, et al. Gemcitabine plus nab-paclitaxel is an active regimen in patients with advanced pancreatic cancer: A phase I/II trial. *J Clin Oncol* 2011;29:4548–4554.

292. Catane R, Von Hoff D, Glaubiger D, Muggia F. Azaserine, DON, and azotomycin: three diazo analogs of L-glutamine with clinical antitumor activity. *Cancer Treat Rep* 1979;63(6):1033–1038.

293. Guo L, Zhou B, Liu Z, Xu Y, Lu H, Xia M. Blockage of glutaminolysis enhances the sensitivity of ovarian cancer cells to PI3K/mTOR inhibition involvement of STAT3 signaling. *Tumor Biol* 2016;37:11007–11015.

294. Han T, Guo M, Zhang T, Gan M, Xie C. A novel glutaminase inhibitor-968 inhibits the migration and proliferation of non-small cell lung cancer cells by targeting EGFR/ERK signaling pathway. *Oncotarget* 2017;8(17):28063–28073.

295. Yuan L, Sheng X, Clark LH, et al. Glutaminase inhibitor compound 968 inhibits cell proliferation and sensitizes paclitaxel in ovarian cancer. *Am J Transl Res* 2016;8(10):4265–4277.

296. Xie C, Jin J, Bao X, Zhan W, Han T, Gan M. Inhibition of mitochondrial glutaminase activity reverses acquired erlotinib resistance in non-small cell lung cancer. *Oncotarget* 2016;7:610–621.

297. Murray-Stewart TR, Woster PM, Casero Jr RA. Targeting polyamine metabolism for cancer therapy and prevention. *Biochem J* 2016;473:2937–2953.

298. Astaldi G, Burgio G., Krc J, Genova R, Astaldi A. J. L-asparaginase and blastogenesis. *Lancet* 1969;1:423.

299. Prager MD, Derr I. Metabolism of asparagine, aspartate, glutamine, and glutamate in lymphoid tissue: Basis for immunosuppression by L-asparaginase. *J Immunol* 1971;106:975–979.

300. Kafkewitz D, Bendich A. Enzyme-induced asparagine and glutamine depletion and immune system function. *Am J Clin Nutr* 1983;37:1025–1030.

301. Torres A, Luke JD, Kullas AL, et al. Asparagine deprivation mediated by Salmonella asparaginase causes suppression of activation-induced T cell metabolic reprogramming. *J Leukoc Biol* 2016;99:387–398.

302. Song P, Wang Z, Zhang X, et al. The role of autophagy in asparaginase-induced immune suppression of macrophages. *Cell Death Dis* 2017;8:e2721.

303. Lucca L, Hafler D. Resisting fatal attraction: a glioma oncometabolite prevents CD8+ T cell recruitment. *J Clin Invest* 2017;127(4):1218–1220.

304. Birendra K, Dinardo CD. Evidence for clinical differentiation and differentiation syndrome in patients with acute myeloid leukemia and IDH1 mutations treated with the targeted mutant IDH1 inhibitor, AG-120. *Clin Lymphoma, Myeloma Leuk* 2016;16(8):460–465.

305. Yen K, Travins J, Wang F, et al. AG-221, a first-in-class therapy targeting acute myeloid leukemia harboring oncogenic IDH2 mutations. *Cancer Discov* 2017;7:478–493.

306. Dang L, Su SM. Isocitrate dehydrogenase mutation and (R)-2-hydroxyglutarate: From basic discovery to therapeutics development. *Annu Rev Biochem* 2017;86:305–331.

307. Yang M, Soga T, Pollard PJ, Adam J. The emerging role of fumarate as an oncometabolite. *Front Oncol* 2012;2:1–7.

308. Mu X, Zhao T, Xu C, Shi W, Geng B, Shen J. Oncometabolite succinate promotes angiogenesis by upregulating VEGF expression through GPR91-mediated STAT3 and ERK activation. *Oncotarget* 2017;8(8):13174–13185.

309. Velaei K, Samadi N, Barazvan B, Soleimani Rad J. Tumor microenvironment-mediated chemoresistance in breast cancer. *Breast* 2016;30:92–100.

310. El Sayed SM, Abou El-Magd RM, Shishido Y, et al. D-Amino acid oxidase-induced oxidative stress, 3-bromopyruvate and citrate inhibit angiogenesis, exhibiting potent anticancer effects. *J Bioenerg Biomembr* 2012;44:513–523.

311. Merchan JR, Kovács K, Railsback JW, et al. Antiangiogenic activity of 2-deoxy-D-glucose. *PLoS One* 2010;5(10):e13699.

312. Roy S, Maity P. Effect of glutamine analogue-acivicin on tumor induced angiogenesis in Ehrlich ascites carcinoma. *Indian J Exp Bol* 2005;43:407–413.

313. Abdel-Aziz AK, Shouman S, El-Demerdash E, Elgendy M, Abdel-Naim AB. Chloroquine synergizes sunitinib cytotoxicity via modulating autophagic, apoptotic and angiogenic machineries. *Chem Biol Interact* 2014;217:28–40.

314. Araújo FA, Rocha MA, Capettini LSA, et al. 3-Hydroxy-3-methylglutaryl coenzyme A reductase inhibitor (fluvastatin) decreases inflammatory angiogenesis in mice. *APMIS* 2013;121:422–430.

315. Kucharzewska P, Welch JE, Svensson KJ, Belting M. Ornithine decarboxylase and extracellular polyamines regulate microvascular sprouting and actin cytoskeleton dynamics in endothelial cells. *Exp Cell Res* 2010;316:2683–2691.

316. Takigawa M, Enomoto M, Nishida Y, Pan HO, Kinoshita A, Suzuki F. Tumor angiogenesis and polyamines: α-difluoromethylornithine, an irreversible inhibitor of ornithine decarboxylase, inhibits B16 melanoma-induced angiogensis in ovo and the proliferation of vascular endothelial cells in vitro. *Cancer Res* 1990;50:4131–4138.

317. Allen E, Miéville P, Warren CM, et al. Metabolic symbiosis enables adaptive resistance to anti-angiogenic therapy that is dependent on mTOR signaling. *Cell Reports* 2016;15:1144-1160

318. Jiménez-Valerio G, Martínez-Lozano M, Bassani N, et al. Resistance to antiangiogenic therapies by metabolic symbiosis in renal cell carcinoma PDX models and patients. *Cell Reports* 2016;15-1134-1143

319. Pisarsky L, Bill R, Fagiani E, et al. Targeting metabolic symbiosis to overcome resistance to anti-angiogenic therapy. *Cell Reports* 2016;15:1161-1174

320. Parra-bonilla G, Alvarez DF, Alexeyev M, Stevens T. Critical role for lactate dehydrogenase A in aerobic glycolysis that sustains pulmonary microvascular endothelial cell proliferation. *Am J Physiol Lung Cell Mol Physiol* 2010;299:513–522.

321. Cantelmo AR, Conradi L-C, Brajic A, et al. Inhibition of the glycolytic activator PFKFB3 in endothelium induces tumor vessel normalization, impairs metastasis, and improves chemotherapy. *Cancer Cell* 2016;30:1–18.

322. Harjes U, Kalucka J, Carmeliet P. Targeting fatty acid metabolism in cancer and endothelial cells. *Crit Rev Oncol Hematol* 2016;97:15–21.

323. Gacche RN, Meshram RJ. Angiogenic factors as potential drug target: Efficacy and limitations of anti-angiogenic therapy. *Biochim Biophys Acta* 2014;1846:161–179.

324. Ramjiawan RR, Griffioen AW, Duda DG. Anti-angiogenesis for cancer revisited: Is there a role for combinations with immunotherapy? *Angiogenesis* 2017;20:185–204.

325. Jiménez-Valerio G, Casanovas O. Angiogenesis and metabolism: entwined for

therapy resistance. *Trends in Cancer* 2017;3(1):10–18.

326. Bueno MJ, Mouron S, Quintela-Fandino M. Personalising and targeting antiangiogenic resistance: A complex and multifactorial approach. *Br J Cancer* 2017;116:1119–1125.

327. Doherty J, Cleveland J. Targeting lactate metabolism for cancer therapeutics. *J Clin Invest* 2013;123(9):3685–3692.

328. Tebbe C, Chhina J, Dar SA, et al. Metformin limits the adipocyte tumor-promoting effect on ovarian cancer. *Oncotarget* 2014;5(13):4746–4764.

329. Martinez-Outschoorn UE, Balliet RM, Rivadeneira DB, et al. Oxidative stress in cancer associated fibroblasts drives tumor-stroma co-evolution: A new paradigm for understanding tumor metabolism, the field effect and genomic instability in cancer cells. *Cell Cycle* 2010;9(16):3256–3276.

330. Romero IL, Mukherjee A, Kenny HA, Litchfield LM, Lengyel E. Molecular pathways: Trafficking of metabolic resources in the tumor microenvironment. *Clin Cancer Res* 2015;21(4):680–686.

331. Allard B, Pommey S, Smyth MJ, Stagg J. Targeting CD73 enhances the antitumor activity of anti-PD-1 and anti-CTLA-4 mAbs. *Clin Cancer Res* 2013;19(20):5626–5635.

332. García-Caballero M, Marí-Beffa M, Medina MA, Quesada AR. Dimethylfumarate inhibits angiogenesis in vitro and in vivo: a possible role for its antipsoriatic effect? *J Invest Dermatol* 2011;131:1347–1355.

333. Eminel S, Jin N, Rostami M, Dibbert S, Mrowietz U, Suhrkamp I. Dimethyl- and monomethylfumarater regulate indolamine 2,3-dioxygenase (IDO) activity in human immune cells. *Exp Dermatol* 2016.

334. Su C, Zhang P, Liu J, Cao Y. Erianin inhibits indoleamine 2,3-dioxygenase-induced tumor angiogenesis. *Biomed Pharmacother* 2017;88:521–528.

335. Panda S. Circadian physiology of metabolism. *Science* 2016;354(6315):317–322.

336. Ullah MS, Davies AJ, Halestrap AP. The plasma membrane lactate transporter MCT4, but not MCT1, is up-regulated by hypoxia through a HIF-1α-dependent mechanism. *J Biol Chem* 2006;281(14):9030–9037.

337. Wakil SJ, Abu-Elheiga LA. Fatty acid metabolism: target for metabolic syndrome. J Lipid Res 2009;50:S138–S143.

338. Rider MH. Role of AMP-activated protein kinase in metabolic depression in animals. *J Comp Physiol B* 2016;186(1):1–16.

339. Draoui N, Feron O. Lactate shuttles at a glance: from physiological paradigms to anti-cancer treatments. *Dis Model Mech* 2011;4:727–732.

340. Skelton MS, Kremer DE, Smith EW, Gladden LB. Lactate influx into red blood cells of athletic and nonathletic species. *Am J Physiol* 1995;268(5):R1121–R1128.

341. Brooks GA. Cell-cell and intracellular lactate shuttles. *J Physiol* 2009;587(23):5591–5600.

342. Pellerin L, Magistretti PJ. Glutamate uptake into astrocytes stimulates aerobic glycolysis: a mechanism coupling neuronal activity to glucose utilization. *Proc Natl Acad Sci* 1994;91:10625–10629.

343. Sieber MH, Spradling AC. The role of metabolic states in development and disease. *Curr Opin Genet Dev* 2017;45:58–68.

344. Hessini K, Kronzucker HJ, Abdelly C, Cruz C. Drought stress obliterates the preference for ammonium as an N source in the C4 plant *Spartina alterniflora*. *J Plant Physiol* 2017;213:98–107.

345. Alkan N, Fortes AM. Insights into molecular and metabolic events associated with fruit response to post-harvest fungal pathogens. *Front Plant Sci* 2015;6:1–14.

346. Zhan T, Digel M, Küch E-M, Stremmel W, Füllekrug J. Silybin and dehydrosilybin decrease glucose uptake by inhibiting GLUT proteins. *J Cell Biochem* 2011;112:849–859.

347. Liu Y, Cao Y, Zhang W, et al. A small-molecule inhibitor of glucose transporter 1 downregulates glycolysis, induces cell-cycle arrest, and inhibits cancer cell growth in vitro and in vivo. *Ther Discov* 2012;11(8):1672–1683.

348. Wood TE, Dalili S, Simpson CD, et al. A novel inhibitor of glucose uptake sensitizes cells to FAS-induced cell death. *Mol Cancer Ther* 2008;7(11):3546–3555.

349. Cao X, Fang L, Gibbs S, et al. Glucose uptake inhibitor sensitizes cancer cells to daunorubicin and overcomes drug resistance in hypoxia. *Cancer Chemother Pharmacol* 2007;59:495–505.

350. Vera JC, Reyes AM, Ca JG, et al. Genistein is a natural inhibitor of hexose and dehydroascorbic acid transport through the glucose transporter, GLUT1. *J Biol Chem* 1996;271(15):8719–8724.

351. Gunnink LK, Alabi OD, Kuiper BD, et al. Curcumin directly inhibits the transport activity of GLUT1. *Biochimie* 2016;125:179–185.

352. Woodward GE, Cramer FB. 2-Desoxy-D-glucose as an inhibitor of anaerobic glycolysis in tumor tissue. *J Franklin Inst* 1952;254(3):259–260.

353. Ko YH, Pedersen PL, Geschwind JF. Glucose catabolism in the rabbit VX2 tumor model for liver cancer: characterization and targeting hexokinase. *Cancer Lett* 2001;173:83–91.

354. Floridi A, Paggi MG, Atri SD, et al. Effect of lonidamine on the energy metabolism of Ehrlich ascites tumor cells. *Cancer Res* 1981;41:4661–4666.

355. Cohen S, Flescher E. Methyl jasmonate: A plant stress hormone as an anti-cancer drug. *Phytochemistry* 2009;70:1600–1609.

356. Clem BF, Neal JO, Tapolsky G, et al. Targeting 6-phosphofructo-2-kinase (PFKFB3) as a therapeutic strategy against cancer. *Mol Cancer Ther* 2013;12(8):1461–1471.

357. McKee RW, Wong W, Landman M. Effects of iodoacetate on glycolysis and respiration in Ehrlich-lettré ascites carcinoma cells. *Biochim Biophys Acta* 1965;105:410–423.

358. Chen J, Xie J, Jiang Z, Wang B, Wang Y, Hu X. Shikonin and its analogs inhibit cancer cell glycolysis by targeting tumor pyruvate kinase-M2. *Oncogene* 2011;30:4297–4306.

359. Le A, Cooper CR, Gouw AM, et al. Inhibition of lactate dehydrogenase A induces oxidative stress and inhibits tumor progression. *Proc Natl Acad Sci* 2010;107(5):2037–2042.

360. Farabegoli F, Vettraino M, Manerba M, Fiume L, Roberti M, Stefano G Di. Galloflavin, a new lactate dehydrogenase inhibitor, induces the death of human breast cancer cells with different glycolytic attitude by affecting distinct signaling pathways. *Eur J Pharm Sci* 2012;47:729–738.

361. Boudreau A, Purkey HE, Hitz A, et al. Metabolic plasticity underpins innate and acquired resistance to LDHA inhibition. *Nat Chem Biol* 2016;12:779–786.

362. Yu Y, Deck JA, Hunsaker LA, et al. Selective active site inhibitors of human lactate dehydrogenases. *Biochem Pharmacol* 2001;62:81–89.

363. Granchi C, Calveresi EC, Tuccinardi T, et al. Assessing the differential action of cancer cells of LDH-A inhibitors based on the N-hydroxyindole-2-carboxylate (NHI) and malonic (Mal) scaffolds. *Org Biomol Chem* 2013;11:6588–6596.

364. Elwood JC. Effect of oxamate on glycolysis and respiration in sarcoma 37 ascites cells. *Cancer Res* 1968;28:2056–2060.

365. Sasaki S, Futagi Y, Ideno M, et al. Effect of diclofenac on SLC16A3/MCT4 by the Caco-2 cell line. *Drug Metab Pharmacokinet* 2016;31(3):218–223.

366. Nancolas B, Guo L, Zhou R, et al. The anti-tumour agent lonidamine is a potent inhibitor of the mitochondrial pyruvate carrier and plasma membrane monocarboxylate transporters. *Biochem J* 2016;473(7):929–936.

367. Baltazar F, Pinheiro C, Queirós O, Preto A, Casal M. Monocarboxylate transporters as targets and mediators in cancer therapy response. *Histol Histopathol* 2014;29:1511–1524.

368. Polanski R, Hodgkinson CL, Fusi A, et al. Activity of the monocarboxylate transporter 1 inhibitor AZD3965 in small cell lung cancer. *Clin Cancer Res* 2014;20(4):926–938.

369. Sonveaux P, Végran F, Schroeder T, et al. Targeting lactate-fueled respiration selectively kills hypoxic tumor cells in mice. *J Clin Invest* 2008;118:3930–3942.

370. Lee KC, Shorr R, Rodriguez R, Maturo C, Boteju LW, Sheldon A. Formation and anti-tumor activity of uncommon in vitro and in vivo metabolites of CPI-613, a novel anti-tumor compound that selectively alters tumor energy metabolism. *Drug Metab Lett* 2011;5:163–182.

371. Papandreou I, Goliasova T, Denko NC. Anticancer drugs that target metabolism: is dichloroacetate the new paradigm? *Int J Cancer* 2011;128:1001–1008.

372. Stacpoole PW, Harman EM, Curry SH, Baumgartner TG, Misbin RI. Treatment of lactic acidosis with dichloroacetate. *N Engl J Med* 1983;309(7):390–396.

373. Stuart SD, Schauble A, Gupta S, et al. A strategically designed small molecule attacks alpha-ketoglutarate dehydrogenase in tumor cells through a redox process. *Cancer Metab* 2014;2:4.

374. Rohle D, Popovici-Muller J, Palaskas N, et al. An inhibitor of mutant IDH1 delays growth and promotes differentiation of glioma cells. *Science* 2013;340(6132):626–630.

375. Wang F, Travins J, DeLaBarre B, et al. Targeted inhibition of mutant IDH2 in leukemia cells induces cellular differentiation. *Science* 2013;340(6132):622–626.

376. Halestrap AP. The mitochondrial pyruvate carrier. Kinetics and specificity for substrates and inhibitors. *Biochem J* 1975;148:85–96.

377. Britten CD, Rowinsky EK, Baker SD, et al. A phase I and pharmacokinetic study of the mitochondrial-specific rhodacyanine dye analog MKT 077. *Clin Cancer Res* 2000;6:42–49.

378. Boukalova S, Stursa J, Werner L, et al. Mitochondrial targeting of metformin enhances its activity against pancreatic cancer. *Mol Cancer Ther* 2016;15(12):2875–2886.

379. Marchiq I, Floch R Le, Simon M, Pouyssegur J. Genetic disruption of lactate/H+ symporters (MCTs) and their subunit CD147/BASIGIN sensitizes glycolytic tumor cells to phenformin. *Cancer Res* 2015;75(1):171–181.

380. Figueras MJ, Gosalvez M. Inhibition of the growth of Ehrlich ascites tumors by treatment with the respiratory inhibitor rotenone. *Eur J Cancer* 1973;9:529–531.

381. Prager R, Schernthaner G. Insulin receptor binding to monocytes, insulin secretion, and glucose tolerance following metformin treatment. Results of a double-blind cross-over study in type II diabetics. *Diabetes* 1983;32:1083–1086.

382. Kroemer G, de Thé H. Arsenic trioxide, a novel mitochondriotoxic anticancer agent? *J Natl Cancer Inst* 1999;91(9):743–745.

383. Seltzer MJ, Bennett BD, Joshi AD, et al. Inhibition of glutaminase preferentially slows growth of glioma cells with mutant IDH1. *Cancer Res* 2010;70(22):8981–8988.

384. Gross MI, Demo SD, Dennison JB, et al. Antitumor activity of the glutaminase inhibitor CB-839 in triple-negative breast cancer. *Mol Cancer Ther* 2014;13:890–901.

385. Altman BJ, Stine ZE, Dang C V. From Krebs to clinic: glutamine metabolism to cancer therapy. *Nat Rev Cancer* 2016;16:619–634.

386. Zhang J, Wang G, Mao Q, et al. Glutamate dehydrogenase (GDH) regulates bioenergetics and redox homeostasis in human glioma. *Oncotarget* 2016;1–12.

387. Thornburg JM, Nelson KK, Clem BF, et al. Targeting aspartate aminotransferase in breast cancer. *Breast Cancer Res* 2008;10:R84.

388. Guth PS, Risey J, Briner W, et al. Evaluation of amino-oxyacetic acid as a palliative in tinnitus. *Ann Otol Rhinol Laryngol* 1990;99(1):74–79.

389. Berge K, Tronstad KJ, Bohov P, Madsen L, Berge RK. Impact of mitochondrial β-oxidation in fatty acid-mediated inhibition of glioma cell proliferation. *J Lipid Res* 2003;44:118–127.

390. Flaig TW, Salzmann-sullivan M, Su L, et al. Lipid catabolism inhibition sensitizes prostate cancer cells to antiandrogen blockade. *Oncotarget* 2017

391. Roberts LN, Mason GP. Clinical trial of a new antianginal drug: Perhexiline maleate. *J Clin Pharmacol* 1972;12(8):342–348.

392. Jones SF, Infante JR. Molecular pathways: Fatty acid synthase. *Clin Cancer Res* 2015;21(24):5434–5439.

393. Pizer ES, Chrest FJ, DiGiuseppe JA, Han WF. Pharmacological inhibitors of mammalian fatty acid synthase suppress DNA replication and induce apoptosis in tumor cell lines. *Cancer Res* 1998;58:4611–4615.

394. Kridel SJ, Axelrod F, Rozenkrantz N, Smith JW. Orlistat is a novel inhibitor of fatty acid synthase with antitumor activity. *Cancer Res* 2004;64:2070–2075.

395. Hanai J, Doro N, Seth P, Sukhatme VP. ATP citrate lyase knockdown impacts cancer stem cells in vitro. *Cell Death Dis* 2013;4:e696.

396. Hatzivassiliou G, Zhao F, Bauer DE, et al. ATP citrate lyase inhibition can suppress tumor cell growth. *Cancer Cell* 2005;8:311–321.

397. Zhou W, Simpson PJ, McFadden JM, et al. Fatty acid synthase inhibition triggers apoptosis during S phase in human cancer cells. *Cancer Res* 2003;63:7330–7337.

398. Clem BF, Clem AL, Yalcin A, et al. A novel small molecule antagonist of choline kinase-α that simultaneously suppresses MAPK and PI3K/AKT signaling. Oncogene 2011;30:3370–3380.

399. Rodríguez-González A, Ramírez de Molina A, Fernández F, et al. Inhibition of choline kinase as a specific cytotoxic strategy in oncogene-transformed cells. *Oncogene* 2003;22:8803–8812.

400. Sánchez-López E, Zimmerman T, Gómez del Pulgar T, Moyer MP, Lacal Sanjuan JC, Cebrian A. Choline kinase inhibition induces exacerbated endoplasmic reticulum stress and triggers apoptosis via CHOP in cancer cells. *Cell Death Dis* 2013;4:e933.

401. de la Cueva A, Ramírez de Molina A, Álvarez-Ayerza N, et al. Combined 5-FU and ChoKα inhibitors as a new alternative therapy of colorectal cancer: evidence in human tumor-derived cell lines and mouse xenografts. *PLoS One* 2013;8(6):e64961.

402. Mashima T, Oh-hara T, Sato S, et al. p53-defective tumors with a functional apoptosome-mediated pathway: A new therapeutic target. *J Natl Cancer Inst* 2005;97(10):765–777.

403. Matusewicz L, Meissner J, Toporkiewicz M, Sikorski AF. The effect of statins on cancer cells — review. *Tumor Biol* 2015;36(7):4889–4904.

404. Kubatka P, Kruzliak P, Rotrekl V, Jelinkova S, Mladosievicova B. Statins in oncological research: From experimental studies to clinical practice. *Crit Rev Oncol Hematol* 2014;92:296–311.

405. Chen Y, Xu Q, Ji D, et al. Inhibition of pentose phosphate pathway suppresses acute myelogenous leukemia. *Tumor Biol* 2016;37(5):6027–6034.

406. Ghashghaeinia M, Giustarini D, Koralkov P, et al. Pharmacological targeting of glucose-6-phosphate dehydrogenase in human erythrocytes by Bay 11–7082, parthenolide and dimethyl fumarate. *Sci Rep* 2016;6:28754.

407. Shin ES, Park J, Shin JM, et al. Catechin gallates are NADP+-competitive inhibitors of glucose-6-phosphate dehydrogenase and other enzymes that employ NADP+ as a coenzyme. *Bioorg Med Chem* 2008;16:3580–3586.

408. Boros LG, Puigjaner J, Cascante M, et al. Oxythiamine and dehydroepiandrosterone inhibit the nonoxidative synthesis of ribose and tumor cell proliferation. *Cancer Res* 1997;57:4242–4248.

409. Hitosugi T, Zhou L, Elf S, et al. Phosphoglycerate mutase 1 coordinates glycolysis and biosynthesis to promote tumor growth. *Cancer Cell* 2012;22:585–600.

410. Feun LG, Kuo MT, Savaraj N. Arginine deprivation in cancer therapy. *Curr Opin Clin Nutr Metab Care* 2015;18:78–82.

411. Yau T, Cheng PN, Chan P, et al. A phase 1 dose-escalating study of pegylated recombinant human arginase 1 (Peg-rhArg1) in patients with advanced hepatocellular carcinoma. *Invest New Drugs* 2013;31:99–107.

412. Lukey MJ, Katt WP, Cerione RA. Targeting amino acid metabolism for cancer therapy. *Drug Discov Today* 2017;22(5):796–804.

413. Serafini P, Meckel K, Kelso M, et al. Phosphodiesterase-5 inhibition augments endogenous antitumor immunity by reducing myeloid-derived suppressor cell function. *J Exp Med* 2006;203(12):2691–2702.

414. Jochems C, Fantini M, Fernando RI, et al. The IDO1 selective inhibitor epacadostat enhances dendritic cell immunogenicity and lytic ability of tumor antigen-specific T cells. *Oncotarget* 2016;7(25):37762–37772.

415. Löb S, Königsrainer A, Rammensee H, Opelz G, Terness P. Inhibitors of indoleamine-2,3-dioxygenase for cancer therapy: can we see the wood for the trees? *Nat Rev Cancer* 2009;9:445–452.

416. Liu X, Shin N, Koblish HK, et al. Selective inhibition of IDO1 effectively regulates mediators of antitumor immunity. *Blood* 2010;115(17):3520–3531.

417. Williams-Ashman HG, Schenone A. Methyl glyoxal bis(guanylhydrazone) as a potent inhibitor of mammalian and yeast S-adenosylmethionine decarboxylases. *Biochem Biophys Res Commun* 1972;46(1):288–295.

418. Siu LL, Rowinsky EK, Hammond LA, et al. A phase I and pharmacokinetic study of SAM486A, a novel polyamine biosynthesis inhibitor, administered on a daily-times-five every-three-week schedule in patients with advanced solid malignancies. *Clin Cancer Res* 2002;8:2157–2166.

419. Samal K, Zhao P, Kendzicky A, et al. AMXT-1501, a novel polyamine transport inhibitor, synergizes with DFMO in inhibiting neuroblastoma cell proliferation by targeting both ornithine decarboxylase and polyamine transport. *Int J Cancer* 2013;133:1323–1334.

420. Tang K-C, Pegg E, Coward JK. Specific and potent inhibition of spermidine synthase by the transitio-state analog, S-adenosyl-3-thio-1,8-diaminooctane. *Biochem Biophys Res Commun* 1980;96(3):1371–1377.

421. Woster PM, Black AY, Duff KJ, Coward JK, Pegg AE. Synthesis and biological evaluation of S-adenosyl-1,12-diamino-3-thio-9-azadodecane, a multisubstrate adduct inhibitor of spermine synthase. *J Med Chem* 1989;32(6):1300–1307.

422. Gonen N, Assaraf YG. Antifolates in cancer therapy: Structure, activity and mechanisms of drug resistance. *Drug Resist Updat* 2012;15:183–210.

423. Wilson PM, Danenberg P V, Johnston PG, Lenz H, Ladner RD. Standing the test of time: targeting thymidylate biosynthesis in cancer therapy. *Nat Rev Clin Oncol* 2014;11:282–298.

424. Ma J, Wang S, Zhao M, et al. Therapeutic potential of cladribine in combination with STAT3 inhibitor against multiple myeloma. *BMC Cancer* 2011;11:255.

425. Cavalcante LDS, Monteiro G. Gemcitabine: Metabolism and molecular mechanisms of action, sensitivity and chemoresistance in pancreatic cancer. *Eur J Pharmacol* 2014;741:8–16.

426. Madaan K, Kaushik D, Verma T. Hydroxyurea: a key player in cancer chemotherapy. *Expert Rev Anticancer Ther* 2012;12(1):19–29.

427. McLaughlin B, Im A, Raptis A, et al. Fludarabine and cytarabine in patients with relapsed acute myeloid leukemia refractory to initial salvage therapy. *Int J Hematol* 2012;96:743–747.

428. Talekar M, Boreddy SR, Singh A, Amiji M. Tumor aerobic glycolysis: new insights into therapeutic strategies with targeted delivery. *Expert Opin Biol Ther* 2014;14(8):1145–1159.

429. Chiarini F, Evangelisti C, Mccubrey JA, Martelli AM. Current treatment strategies for inhibiting mTOR in cancer. *Trends Pharmacol Sci* 2015;36(2):124–135.

430. Maráz A, Csejtei A, Kocsis J, et al. Assessment of the role of everolimus therapy in patients with renal cell carcinoma based on daily routine and recent research results. *Pathol Oncol Res* 2017.

431. Dasgupta S, Rajapakshe K, Zhu B, et al. Metabolic enzyme PFKFB4 activates transcriptional coactivator SRC-3 to drive breast cancer. *Nature* 2018;556:249-254

432. Garber K. First metabolic oncology inhibitor gets FDA green light, with record price tag. *Nat Biotech* 2017;35:895

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Beatriz Martínez Poveda was graduated in Biology at the University of Malaga (Spain) in 2002 and she achieved her international PhD at the same University in 2007 working on characterization of new natural compounds with anti-angiogenic potential. Then she moved to Madrid for a first post-doctoral period in the Biomedical Research Institute (IIB, Madrid), focusing on the study of hypoxia and anti-angiogenic therapy in tumors using *in vivo* imaging techniques. In 2009, she started a second post-doctoral period in the Cardiovascular Research National Institute (CNIC, Madrid) working mainly in the molecular characterization of cardiovascular diseases, with significant contributions in the field of aortic valve stenosis and calcification, atherosclerosis and left ventricle non-compaction. Since 2015 Dr. Martínez-Poveda is working as Assistant Professor in the Department of Molecular Biology and Biochemistry at the University of Málaga, and as a post-doctoral researcher in projects related to tumoral angiogenesis and inflammation.

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Table 1. Metabolic modulators with proved anti-tumor activity.

Target	Drug	Observations	References
Glycolysis			
GLUT1	Curcumin Fasentin Genistein Phloretin Silibinin WZB117	Silibinin is in Phase II of clinical trials (prostate cancer).[a] Curcumin[b] and genistein[c] are in clinical trials (multiple kinds of cancer).	346-351
Hexokinases	2-DG 3-bromopyruvate Lonidamine Methyl jasmonate	Lonidamine is in Phase III of clinical trials (prostate cancer).[d] 2-DG is in clinical trials (multiple kinds of cancer).[e]	352-355
PFKFB3	3PO PFK15		321,356
G3PDH	Iodoacetate		357
PKM2	Shikonin		358
LDH-A	FX11 Galloflavin GNE-140 Gossypol NHI Oxamate Panepoxydone	Gossypol is in clinical trials (multiple kinds of cancer).[f]	20,359–364
Lactate secretion			
MCT4	Diclofenac Lonidamine	Diclofenac is FDA approved (anti-inflammatory drug). Lonidamine is in Phase III of clinical trials (prostate cancer).[d]	365,366
Lactate uptake			
MCT1	AR-C155858 AZD3965 CHC	AR-C155858 is in preclinical studies. AZD3965 is in Phase I of	366–369

	Lonidamine	clinical trials (gastric cancer, prostate cancer and lymphoma).[g] Lonidamine is in Phase III of clinical trials (prostate cancer).[d]	
TCA cycle			
PDH	CPI-613	Clinical trials (multiple kinds of cancer).[h]	370
PDK1	DCA	Approved for the treatment of lactic acidosis.	371,372
KGDH	CPI-613	Clinical trials (multiple kinds of cancer).[h]	373
IDH	AG-120 (ivosidenib) AG-221 (enasidenib) AGI-5198 AGI-6780	Ivosidenib[i] and enasidenib[j] are in Phase III of clinical trials (leukemia).*	304,305,374,375
MPC	Lonidamine UK-5099	Lonidamine is in Phase III of clinical trials (prostate cancer).[d]	366,376
OXPHOS			
Mitochondrial potential membrane	MKT-077		377
Mitochondrial complex I	Metformin Phenformin Rotenone	Metformin is approved for the treatment of type 2 diabetes. Phenformin is in Phase I of clinical trials (melanoma).[k]	378–381
Mitochondrial complex III	Arsenic trioxide	FDA approved for the treatment of acute promyelocytic leukemia.	382
Glutamine metabolism			
Glutamine antimetabolite	Acivicin Azaserine DON	Not approved for clinical due to toxicity.	12
GLS1	968 BPTES CB-839	CB-839 is in clinical trials (multiple kinds of cancer).[l]	293–296,383,384

SLC1A5	Benzylserine γ-FBP GPNA		[385]
GLUD	EGCG R162	EGCG is in clinical trials (multiple kinds of cancer).[m]	[386]
Aminotransferases	AOA	Approved for the treatment of tinnitus.	[387,388]
Fatty acid β-oxidation			
CPT1	Aminocarnitine Etomoxir Perhexiline Ranolazine	Perhexiline and ranolazine are approved for use as an anti-angina therapy.	[389–391]
Lipid synthesis			
FAS	C75 Cerulenin Orlistat TVB-2640	Orlistat is approved for the treatment of obesity. TVB-2640 is in Phase II of clinical trials (multiple kinds of cancer).[n]	[392–394]
ACL	Hydroxycitrate SB-204990		[395,396]
ACC	TOFA		[397]
Choline kinase	CK37 MN58b RSM932A TCD-717	TCD-717 is in Phase I of clinical trials (advanced solid tumors).[o]	[398–401]
ACS	Triacsin C		[402]
Mevalonate pathway			
HMGCR	Statins	Approved for the treatment of hypercholesterolaemia	[403,404]
Pentose phosphate pathway			
G6PDH	6-aminonicotinamide DHEA DMF EGCG	EGCG is in clinical trials (multiple kinds of cancer).[m] Dimethylfumarate is FDA approved (multiple	[405–408]

		sclerosis).	
PGAM1	PGMI-004A		[409]
Amino acid metabolism			
Asparagine availability	L-asparaginase	FDA approved for the treatment of acute lymphoblastic leukemia, acute myeloid leukemia, and non-Hodgkin's lymphoma.	[58,63]
Arginine availability	Pegylated arginine deiminase (ADI-PEG20) rhArg1-PEG (BCT-100)	BCT-100 is in Phase II of clinical trials (multiple kinds of cancer).[p] ADI-PEG20 is in clinical trials (multiple kinds of cancer).[q]	[410–412]
Arginase	Tadalafil (Cialis)	FDA approved for the treatment of benign prostatic hypertrophy.	[14,413]
IDO	1-methyl-trytophan (Indoximod) DMF Epacadostat Erianin	Indoximod[r] and epacadostat[s] are in clinical trials (multiple kinds of cancer). Dimethylfumarate is FDA approved (multiple sclerosis).	[333,334,414–416]
Polyamine metabolism			
ODC	DFMO	Phase II of clinical trials (neuroblastoma).[t]	[84]
AMD1	MGBG SAM486A	MGBG is toxic for clinical development.	[85,417,418]
Polyamine transport	AMXT-1501	.	[419]
Aminopropyltrans-ferases	AdoDATAD AdoDATO		[420,421]
Polyamine analogs	BENSpm CPENSpm PG-11047 PG-11093	PG-11047 is in Phase I of clinical trials (advanced refractory solid tumors and lymphoma).[u]	[297]
Nucleid acid synthesis			

DHFR	Methotrexate Pemetrexed Pralatrexate Trimitrexate (antifolates)	Methotrexate is FDA approved for treatment of cancer, autoimmune diseases, ectopic pregnancy, and for medical abortions. Pemetrexed is FDA approved for the treatment of pleural mesothelioma and non-small cell lung cancer. Pralatrexate is FDA approved relapsed or refractory peripheral T-cell lymphoma.	[422]
Thymidylate synthase	5-fluorouracil Raltitrexed	5-fluorouracil is FDA approved for the treatment of several kinds of cancer. Raltitrexed is in Phase IV of clinical trials (multiple kinds of cancer).[v]	[423]
Adenine/adenosine deaminase	Cladribine	FDA approved for the treatment of hairy cell leukemia and B-cell chronic lymphocytic leukemia.	[424]
DNA polymerase/ ribonucleotide reductase	Cytarabine Fludarabine Gemcitabine Hydroxyurea	Cytarabine is FDA approved for the treatment of acute myeloid leukemia, acute lymphocytic leukemia, chronic myelogenous leukemia, and non-Hodgkin's lymphoma. Fludarabine is FDA approved for the treatment of leukemia and lymphoma. Gemcitabine is FDA approved for the treatment of several kinds of cancer.	[425–427]

		Hydroxyurea is FDS approved for the treatment of sickle-cell disease, chronic myelogenous leukemia, cervical cancer, and polycythemia vera.	
Nitric oxide metabolism			
NOS	L-NAME		[190]
Metabolic signaling pathways			
HIF-1	Digoxin Irinotecan PX478 Topotecan	PX478 is in Phase I of clinical trials (advanced solid tumors and lymphoma).[w] Digoxin is FDA approved for the treatment of several heart diseases. Irinotecan is FDA approved for the treatment of colon and small cell lung cancer. Topotecan is FDA approved for the treatment of several kinds of cancer.	[428]
mTOR	Everolimus PP242 Temsirolimus	Everolimus and temsirolimus are also approved immunosuppressants. Everolimus is approved for the treatment of advanced kidney cancer.	[293,429,430]

2-DG, 2-deoxyglucose; ACC, acetyl-CoA carboxylase; ACL, ATP citrate lyase; ACS, acyl-CoA synthetase; AdoDATAD, S-adenosyl-1,12-diamino-3-thio-9-azadodecane; AdoDATO, S-adenosyl-3-thio-1,8-diaminooctane; AMD1, adenosylmethionine decarboxylase; AOA, aminooxyacetate; BPTES, bis-2-(5-phenylacetamido-1,3,4-thiadiazol-2-yl)ethyl sulfide; CHC, α-cyano-4-hydroxycinnamic acid; CPT1, carnitine palmitoyltransferase 1; DCA, dichloroacetate; DFMO, difluoromethylornithine; DHEA,

dehydroepiandrosterone; DHFR, dihydrofolate reductase; DMF, dimethylfumarate; DON, 6-diazo-5-oxo-L-norleucine; EGCG, epigallocatechin gallate; FAS, fatty acid synthase; γ-FBP, γ-folate binding protein; G3PDH, glyceraldehyde-3-phosphate dehydrogenase; GLS1, glutaminase; G6PDH, glucose-6-phosphate dehydrogenase; GPNA, L-γ-glutamyl-p-nitroanilide; GLUD, glutamate dehydrogenase; HIF-1, hypoxia-inducible factor 1; HMGCR, HMG-CoA reductase; IDH, isocitrate dehydrogenases; IDO, indoleamine-2,3-dioxygenase; KGDH, α-ketoglutarate dehydrogenase; LDH-A, lactate dehydrogenase A; L-NAME, L-NG-nitroarginine methyl ester; MGBG, methylglyoxal(bis)guanylhydrazone; MPC, mitochondrial pyruvate carrier; mTOR, mammalian target of rapamycin; NHI, N-hydroxy-2-carboxy-substituted indoles; NOS, nitric oxide synthase; ODC, ornithine decarboxylase; OXPHOS, oxidative phosphorylation; PDH, pyruvate dehydrogenase; PDK1, pyruvate dehydrogenase kinase 1; PFKFB3, phosphofructokinase-2/fructose-2,6-bisphosphatase 3; PGAM1, phosphoglycerate mutase; PKM2, pyruvate kinase M2; TCA, tricarboxylic acid cycle.
[a]ClinicalTrials.gov Identifier: NCT00487721; [b]pancreatic cancer, phase II, ClinicalTrials.gov Identifier: NCT00192842; breast cancer, phase II, ClinicalTrials.gov Identifier: NCT01042938; endometrial carcinoma, phase II, ClinicalTrials.gov Identifier: NCT02017353; head and neck cancer, early phase I, ClinicalTrials.gov Identifier: NCT01160302; pancreatic cancer, phase II, ClinicalTrials.gov Identifier: NCT00094445; colorectal cancer, phase I, ClinicalTrials.gov Identifier: NCT00027495; multiple myeloma, ClinicalTrials.gov Identifier: NCT00113841; prostate cancer, phase III, ClinicalTrials.gov Identifier: NCT02064673; osteosarcoma, phase II, ClinicalTrials.gov Identifier: NCT00689195; [c]prostate cancer, phase III, ClinicalTrials.gov Identifier: NCT00584532; kidney cancer and melanoma, early phase I, ClinicalTrials.gov Identifier: NCT00276835; breast cancer, phase II, ClinicalTrials.gov Identifier: NCT00244933; bladder cancer, phase II, ClinicalTrials.gov Identifier: NCT00118040; non small cell lung cancer, phase II, ClinicalTrials.gov Identifier: NCT01628471; pancreatic cancer, phase II, ClinicalTrials.gov Identifier: NCT00376948; colorectal cancer, phase II, ClinicalTrials.gov Identifier: NCT01985763; [d]ClinicalTrials.gov Identifier: NCT00435448; [e]prostate cancer, phase II, ClinicalTrials.gov Identifier: NCT00633087; lung cancer, breast cancer, pancreatic cancer, gastric cancer and head and neck cancer, phase I, ClinicalTrials.gov Identifier: NCT00096707; [f]adult glioblastoma, phase II, ClinicalTrials.gov Identifier: NCT00540722; lymphoma, phase II, ClinicalTrials.gov Identifier: NCT00275431; adrenocortical carcinoma, phase II, ClinicalTrials.gov Identifier: NCT00848016; leukemia, phase II, ClinicalTrials.gov Identifier: NCT00286780; laryngeal cancer, phase II, ClinicalTrials.gov Identifier: NCT01633541; small cell lung cancer, phase II, ClinicalTrials.gov Identifier: NCT00773955; prostate cancer, phase II, ClinicalTrials.gov Identifier: NCT00666666; [g]ClinicalTrials.gov Identifier: NCT01791595; [h]small cell lung cancer, phase I, ClinicalTrials.gov Identifier: NCT01931787; pancreatic cancer, phase I, ClinicalTrials.gov Identifier: NCT01839981; colorectal cancer, phase I, ClinicalTrials.gov Identifier: NCT02232152; adult acute myeloid leukemia, phase I, ClinicalTrials.gov Identifier: NCT01768897; lymphoma, phase I, ClinicalTrials.gov Identifier: NCT02168140; [i]ClinicalTrials.gov Identifier:

NCT03173248; [j]ClinicalTrials.gov Identifier: NCT02577406; [k]ClinicalTrials.gov Identifier: NCT03026517; [l]colorectal cancer, phase II, ClinicalTrials.gov Identifier: NCT02861300; lymphoma, phase I, ClinicalTrials.gov Identifier: NCT02071888; leukemia, phase I, ClinicalTrials.gov Identifier: NCT02071927; breast cancer, phase II, ClinicalTrials.gov Identifier: NCT03057600; renal cell carcinoma, phase II, ClinicalTrials.gov Identifier: NCT03428217; [m]colon cancer, early phase I, ClinicalTrials.gov Identifier: NCT02891538; bladder cancer, phase II, ClinicalTrials.gov Identifier: NCT00666562; breast cancer, phase II, ClinicalTrials.gov Identifier: NCT00917735; prostate cancer, phase II, ClinicalTrials.gov Identifier: NCT00676780; [n]breast cancer, phase II, ClinicalTrials.gov Identifier: NCT03179904; colon cancer, phase I, ClinicalTrials.gov Identifier: NCT02980029; astrocytoma, phase II, ClinicalTrials.gov Identifier: NCT03032484; [o]ClinicalTrials.gov Identifier: NCT01215864; [p]hepatocellular carcinoma, phase II, ClinicalTrials.gov Identifier: NCT01092091; leukemia, phase II, ClinicalTrials.gov Identifier: NCT02899286; renal cell carcinoma, melanoma and prostate adenocarcinoma, phase I, ClinicalTrials.gov Identifier: NCT02285101; [q]melanoma, phase II, ClinicalTrials.gov Identifier: NCT00520299; prostate cancer, phase I, ClinicalTrials.gov Identifier: NCT01497925; breast cancer, phase I, ClinicalTrials.gov Identifier: NCT01948843; acute myeloid leukemia, phase I, ClinicalTrials.gov Identifier: NCT02875093; hepatocellular carcinoma, phase III, ClinicalTrials.gov Identifier: NCT01287585; [r]glioblastoma, phase II, ClinicalTrials.gov Identifier: NCT02052648; pancreatic cancer, phase II, ClinicalTrials.gov Identifier: NCT02077881; prostate cancer, phase II, ClinicalTrials.gov Identifier: NCT01560923; melanoma, phase III, ClinicalTrials.gov Identifier: NCT03301636; acute myeloid leukemia, phase II, ClinicalTrials.gov Identifier: NCT02835729; [s]sarcoma, phase II, ClinicalTrials.gov Identifier: NCT03414229; lymphoma and solid tumors, phase II, ClinicalTrials.gov Identifier: NCT03322384; renal cell carcinoma, phase III, ClinicalTrials.gov Identifier: NCT03260894; urothelial cancer, phase III, ClinicalTrials.gov Identifier: NCT03374488; head and neck cancer, phase III, ClinicalTrials.gov Identifier: NCT03342352; lung cancer, phase III, ClinicalTrials.gov Identifier: NCT03322566; pancreatic cancer, phase II, ClinicalTrials.gov Identifier: NCT03006302; prostate cancer, phase II, ClinicalTrials.gov Identifier: NCT03493945; ovarian cancer, phase I, ClinicalTrials.gov Identifier: NCT02118285; [t]ClinicalTrials.gov Identifier: NCT02679144; [u]advanced refractory solid tumors, phase I, ClinicalTrials.gov Identifier: NCT00705653; lymphoma, phase I, ClinicalTrials.gov Identifier: NCT00293488; [v]head and neck cancer, phase IV, ClinicalTrials.gov Identifier: NCT03196843; nasopharyngeal carcinoma, phase II, ClinicalTrials.gov Identifier: NCT02562599; childhood leukemia, phase I, ClinicalTrials.gov Identifier: NCT00003528; gastric cancer, phase II, ClinicalTrials.gov Identifier: NCT03392103; colorectal cancer, phase IV, ClinicalTrials.gov Identifier: NCT01959061; [w]ClinicalTrials.gov Identifier: NCT00522652; *enasidenib has already been approved by FDA (see Notes added in proof).

FIGURE CAPTIONS

Figure 1. Important aspects regarding metabolism of tumor cells and several cells of the tumor microenvironment.

Figure 2. Role of different cells of the tumor microenvironment in immunosuppression. Different cells of the tumor microenvironment are able to affect the immune activity. Proliferation of Treg cells is modulated by TGF-β from cancer-associated fibroblasts (CAFs) and tumor cells and by IL-10 secreted by tumor-associated macrophages (TAMs). Tumor cells consume high amounts of tryptophan and arginine, thus depleting them from the media. TAMs also consume tryptophan, and HIF-1α induces the expression of arginase 1 (Arg1), hence diminishing arginine concentration in the extracellular media. Part of the arginine consumed by tumor cells can be leaded to nitric oxide (NO) synthesis, which inhibits effector T cells activity. Additionally, the high uptake of glutamine by tumor cells decreases glutamine availability in the media, inhibiting glutaminolysis in effector T cells, which, in turn, impairs polyamine and nucleotide synthesis in these cells. Tumor cells also express CD73 marker, responsible for increasing AMP concentration in the media, which will be converted to adenosine, capable of inhibiting immune response by effector T cells. Regarding glucose metabolism, TAMs and tumor cells express PD-L1, the ligand for PD-1, and their interaction inhibits glycolysis in effector T cells. PD-L1 favors the high glycolytic rate in tumor cells, thus depleting glucose from the media, and then the transcription of IFN-γ and IL-2 is inhibited. All these facts lead to immunosuppression. Solid arrows show production or secretion; dashed arrows represent induction or inhibition; dotdashed arrows indicate a substrate or process integrated to another process; thicker arrows depict a higher rate of incorporation of the indicated substrate.

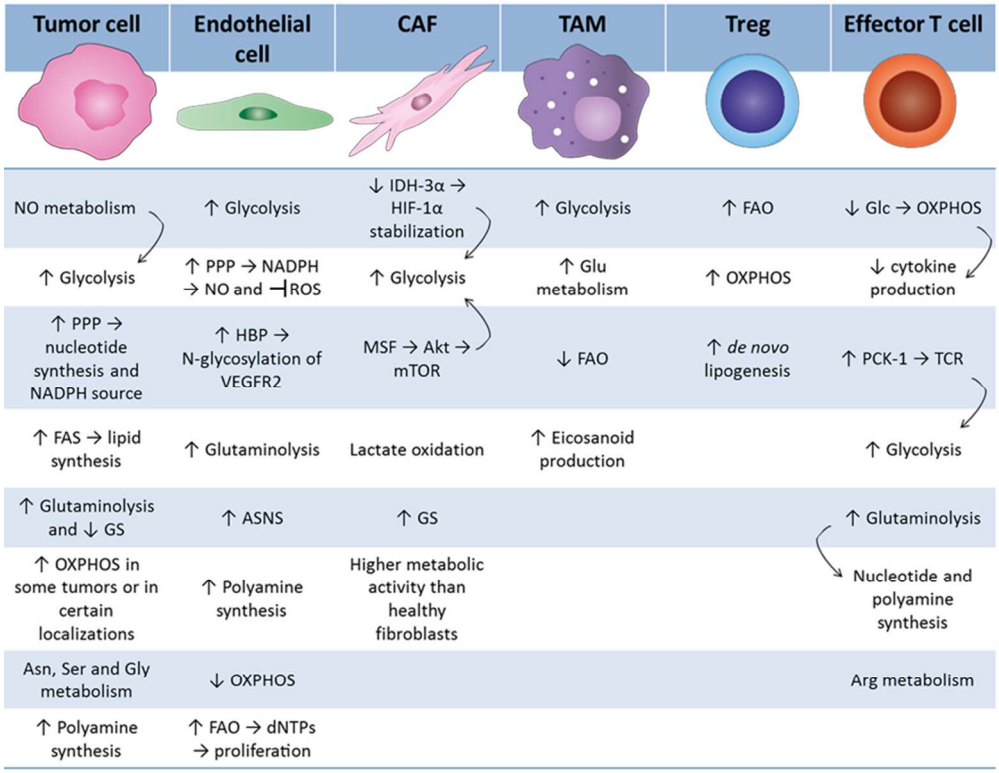
Figure 3. Metabolite exchange between tumor cells and different cells of the tumor microenvironment and its relation with tumor progression. There are multiple metabolic interactions between the different cells of the tumor microenvironment. For example, endothelial cells (ECs) consume lactate produced by tumor cells, thus enhancing the angiogenic process, and ECs extrude mitochondria to tumor cells, conferring them chemoresistance. Lactate from tumor cells are also consumed by tumor-associated macrophages (TAMs) and cancer-associated fibroblasts (CAFs). On the one hand, in TAMs, lactate stabilizes HIF-1α, thus promoting angiogenesis and immunosuppression. On the other hand, in CAFs lactate induces hyaluronic acid production, which contributes to tumor invasiveness along with kynurenine, a tryptophan metabolite produced by tumor cells and TAMs. Lactate production by CAFs is also promoted by ROS liberation from tumor cells. Additionally, cancer-associated adipocytes (CAAs), TAMs and CAFs synthetize glutamine, which is uptaken by tumor cells. CAAs and CAFs also provide fatty acids (FAs) to tumor cells. Moreover, CAAs supply tumor cells with citrulline and arginine, hence contributing to polyamine and nitric oxide (NO) synthesis in these cells. Solid arrows show production or secretion; dashed arrows represent induction or inhibition; dotdashed arrows indicate a substrate or process

integrated to another process.

Figure 4. Role of different cells of the tumor microenvironment in promoting angiogenesis. Tumor cells contribute to activation of angiogenesis through lactate secretion to the media, which is consumed by endothelial cells (ECs). ECs are also able to produce lactate via glycolysis, and this lactate promotes the phosphorylation of Akt, which, in turn, promotes the glycolytic process in a positive feed-back. Indirectly, lactate inhibits prolyl hydroxylases (PHD). PHD inhibition enables stabilization of HIF-1α and the liberation of the active form of NF-κB, thus allowing the transcription of pro-angiogenic factors such as basic fibroblast growth factor (bFGF), vascular endothelial growth factor receptor 2 (VEGFR2), vascular endothelial growth factor (VEGF) and interleukin 8 (IL-8). VEGF, as well, promotes fatty acid (FA) uptake in ECs. Oxidation of these fatty acids leads to nucleotide synthesis, increasing EC proliferation. Moreover, expression of β2-adrenergic receptor (ADRβ2) favors the glycolytic phenotype through inhibition of OXPHOS. Additionally, other cells of the tumor microenvironment are also able to modulate angiogenesis. For example, stabilization of HIF-1α by ROS liberation from tumors increases the glycolytic rate in cancer-associated fibroblasts (CAFs), and the resulting lactate promotes the liberation of metalloproteinase-9 (MMP9) to the media. Furthermore, TGF-β expressed in these cells activates urokinase-type plasminogen activator (uPA). Both molecules are involved in extracellular matrix degradation. On the other hand, tumor-associated macrofages (TAMs) produce TNF-α, which allows the expression of MMP9 and uPA as well, and of IL-1, which upregulates HIF-1α, hence increasing transcription of VEGF and other pro-angiogenic factors. It has to be taken into account that many other factors produced by the different cells of the microenvironment regulate the angiogenic process, but they are not represented here for the sake of clarity. Solid arrows show production or secretion; dashed arrows represent induction or inhibition; dotdashed arrows indicate a substrate or process integrated to another process.

Figure 5. Interactions between tumor and host metabolism. Tumor growth is promoted by means of different metabolic interactions of tumor with host tissues. Tumors secrete IL-6, which has two effects on the liver: i) inhibiting ketogenesis, which stimulates the secretion of adrenocortical hormones (ACH), therefore promoting protein catabolism in muscles, which results in free amino acids for their use by the tumor, and ii) promoting insulin liberation, which induces gluconeogenesis in the liver, thus supplying the tumor with glucose. In addition, gluconeogenesis in the liver also uses alanine from muscles and lactate from muscles and the tumor (all this corresponding to the so-called Cori cycle), and gluconeogenesis is also carried out in the kidneys. Moreover, tumors act as "nitrogen traps", consuming high amounts of glutamine from the blood. Liver and kidneys have a high glutamine synthetase (GS) and a low glutaminase (GLS) expression, and muscles present high GS expression, thus providing tumors with glutamine. This high uptake of glutamine by the tumor decreases glutamine available for natural-killer (NK) cells, thus diminishing glutathione (GSH) concentration and affecting NK cells activity. Tumors also consume arginine, depleting the arginine

available for other tissues. In addition, tumors take up uridine from lympohid organs, leading to a decrease in RNA synthesis in these organs. All this contributes to immunosuppression. The arginine consumed can be used for nitric oxide (NO) and polyamine synthesis, helped by a high uptake of ornithine and methionine from the tissues, as well as a high ornithine decarboxylase (ODC) activity. Besides, lipid catabolism is promoted in the adipose tissue, thus liberating free fatty acids (FAs) to the blood that are uptaken by the tumor. Solid arrows show production or secretion; dashed arrows represent induction or inhibition; dotdashed arrows indicate a substrate or process integrated to another process; thicker arrows depict a higher rate of incorporation of the indicated substrate.



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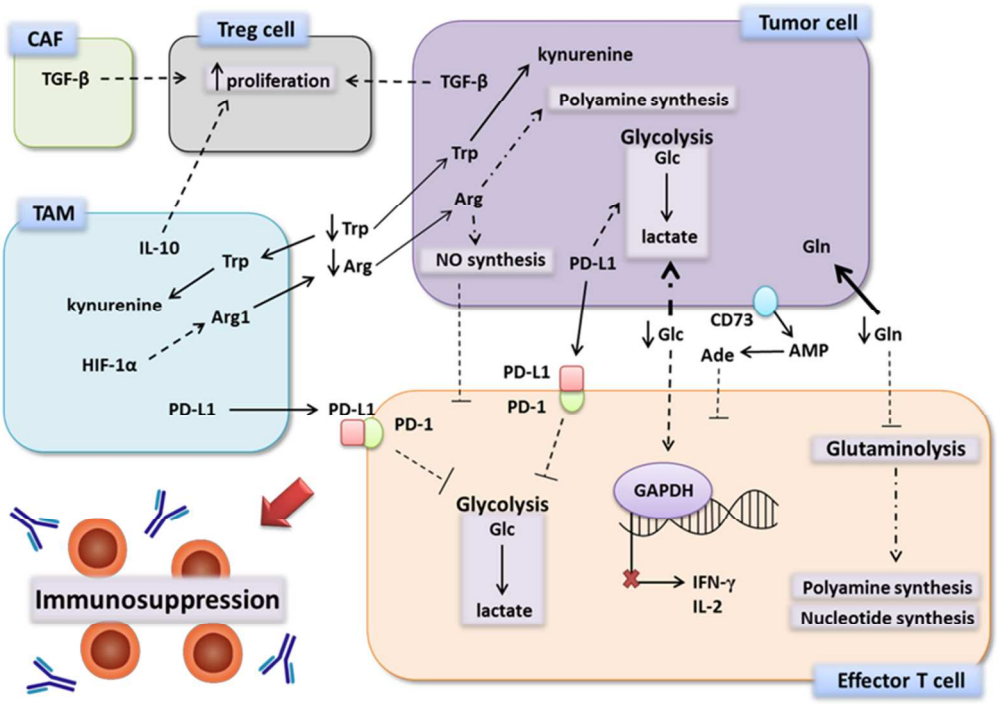

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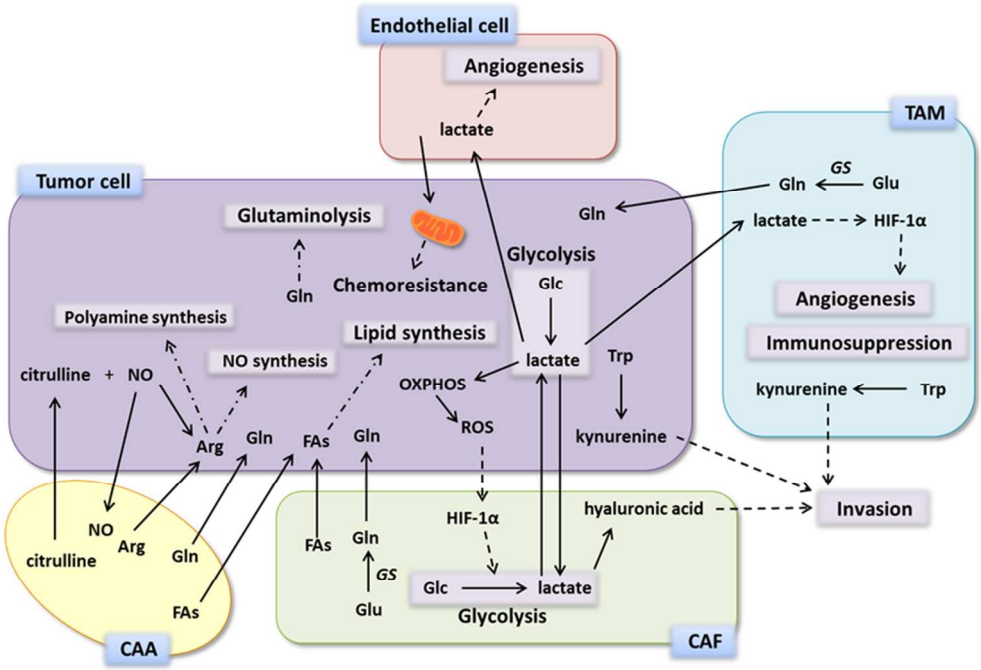

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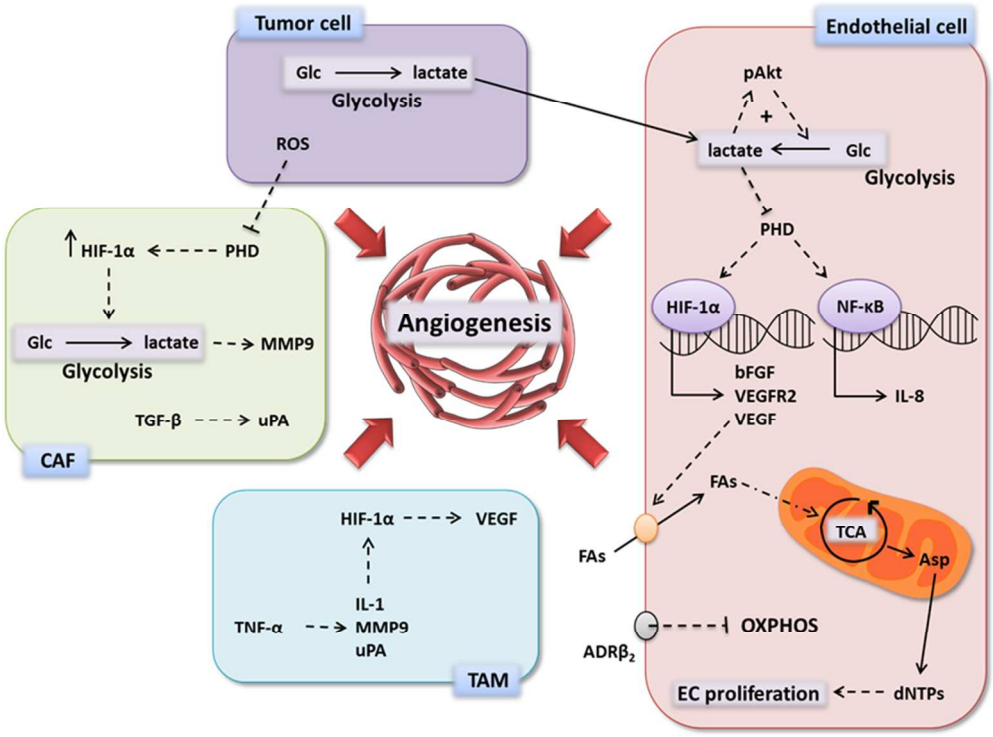

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