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Macrophages and metabolism in the tumor microenvironment

Ilio Vitale^{1,2,3}, Gwenola Manic⁴, Lisa M. Coussens⁵, Guido Kroemer^{6,7,8,9,10,11,12,13,*} and Lorenzo Galluzzi^{6,14,15,16,*}

¹Department of Biology, University of Rome "Tor Vergata", Rome, Italy; ²Italian Institute for Genomic Medicine (IIGM), Turin, Italy; ³Candiolo Cancer Institute - FPO, IRCCS, Candiolo, Italy; ⁴IRCSS - Regina Elena National Cancer Institute, Rome, Italy; ⁵Department of Cell Development & Cancer Biology, Knight Cancer Institute, Oregon Health and Science University, Portland, OR, USA; ⁶Université Paris Descartes, Paris, France; ⁷Université Pierre et Marie Curie, Paris, France; ⁸Equipe 11 labellisée Ligue Nationale contre le Cancer, Centre de Recherche des Cordeliers, Paris, France; ⁹INSERM, U1138; Paris, France; ¹⁰Metabolomics and Cell Biology Platforms, Gustave Roussy Cancer Campus, Villejuif, France; ¹¹Pôle de Biologie, Hôpital Européen Georges Pompidou, AP-HP, Paris, France; ¹²Suzhou Institute for Systems Biology, Chinese Academy of Sciences, Suzhou, China; ¹³Department of Women's and Children's Health, Karolinska University Hospital, Stockholm, Sweden; ¹⁴Department of Radiation Oncology, Weill Cornell Medical College, New York, NY, USA; ¹⁵Sandra and Edward Meyer Cancer Center, New York, NY, USA; ¹⁶Department of Dermatology, Yale University School of Medicine, New Haven, CT, USA.

*Correspondence: deadoc80@gmail.com (L. Galluzzi, lead contact), or kroemer@orange.fr (G. Kroemer).

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Abstract

Tumor-associated macrophages (TAMs) constitute a plastic and heterogeneous cell population of the tumor microenvironment (TME) that can account for up to 50% of some solid neoplasms. Most often, TAMs support disease progression and resistance to therapy by providing malignant cells with trophic and nutritional support. However, TAMs can mediate antineoplastic effects, especially in response to pharmacological agents that boost their phagocytic and oxidative functions. Thus, TAMs and their impact on the overall metabolic profile of the TME have a major influence on tumor progression and resistance to therapy, *de facto* constituting promising targets for the development of novel anticancer agents. Here, we discuss the metabolic circuitries whereby TAMs condition the TME to support tumor growth, and how such pathways can be therapeutically targeted.

Introduction

The homeostasis and evolution of the tumor microenvironment (TME) are governed by an intimate crosstalk within and across all cellular compartments, including malignant, endothelial, stromal and immune cells. Such a complex interaction often involves extracellular metabolites, which not only constitute a source of energy supply, but also act as communication signals between different cellular compartments. As an example, of this metabolic crosstalk, cancer-associated fibroblasts (CAFs) and adipocytes can support malignant cells by providing nutrients such as alanine and lipids into the TME (Nieman et al., 2011; Sousa et al., 2016; Vitale et al., 2019; Zhang et al., 2018a). Moreover, cancer cells can harness metabolic byproducts to hijack the functions of tumor-infiltrating immune cells to their own benefit. This applies to lactate secreted by glycolytic cancer cells, which often favors the polarization of immune cells to an immunosuppressive phenotypes (Angelin et al., 2017; Chen et al., 2017a; Colegio et al., 2014). Finally, all cells of the TME compete with each other for nutrients and oxygen, both of which are generally limited (Bantug et al., 2018; Chang et al., 2015). All these nutritional constraints shape the metabolism of evolving neoplasms, hence acting as a prominent evolutionary force (Buck et al., 2017).

Tumor-associated macrophages (TAMs) and their precursors account for the largest fraction of the myeloid infiltrate in the majority of human solid malignancies, as shown by immunohistochemical analyses of TAM markers such as CD68, as well as by CIBERSORT-mediated dissection of gene expression profiles (although with reduced specificity) (Cassetta et al., 2019; Chevrier et al., 2017; Gentles et al., 2015; Wagner et al., 2019). The TAM compartment is highly dynamic and heterogeneous (**Box 1**), both within and across tumors (Chevrier et al., 2017; Cuccarese et al., 2017; Lavin et al., 2017). An elevated degree of TAM heterogeneity has indeed been revealed not only across different cancer patients, but also across different malignant lesions of the same patient, as well as

within a specific neoplastic lesion. Part of such heterogeneity reflects the ability of TAMs to acquire an entire spectrum of phenotypic, metabolic and functional profiles ranging from a pro-inflammatory (so-called M1-like) to an anti-inflammatory (so-called M2-like) state in response to environmental perturbations (Cassetta and Pollard, 2018; Mills et al., 2000). Thus, while specific TAM subsets support oncogenesis, vascularization, disease progression in spite of immunosurveillance (immuno-evasion), and resistance to treatment, ultimately correlating with poor disease outcome (Gentles et al., 2015; Mantovani et al., 2017; Wenes et al., 2016), other TAM populations exert tumoricidal functions and support (rather than counteract) the efficacy of various anticancer (immuno)therapies (Arnold et al., 2014; Mantovani et al., 2017). Thus, TAMs stand out as obstacles – and hence promising targets – for the development of novel anticancer immunotherapies (Cassetta and Pollard, 2018; Ruffell and Coussens, 2015). Of note, at least in some tumors including glioblastoma and pancreatic cancer, TAMs can derive from both tissue-resident macrophages and circulating monocytes, and TAM ontogeny appears to influence their functional profile (Bowman et al., 2016; Chen et al., 2017b; Loyher et al., 2018; Zhu et al., 2017). Moreover, the precise localization of TAMs within the TME directs (at least in part) their ability to support *versus* restrain tumor progression. Specifically, TAM subpopulations TAMs at perivascular or hypoxic areas reportedly display proangiogenic and immunosuppressive properties (Coffelt et al., 2010; Laoui et al., 2014), while (at least in some settings) TAMs populating the invasive tumor front or cancer cell nests exhibit tumoricidal activity (Forsell et al., 2007; Yang et al., 2018).

Here, we present the major metabolic circuitries that underlie the ability of TAMs to shape the TME of developing tumors, discuss the metabolic cues of the TME that influence functional TAM polarization, and propose immunometabolic strategies to harness TAMs for tumor prevention and treatment.

TAM metabolism in tumor progression

The TAM compartment evolves over time (*i.e.*, during tumor progression and response to treatment) as well as in space (*i.e.*, at different tumor sites) (**Box 2**) through an extensive remodeling of core energy metabolism (Mazzone et al., 2018). M1-like macrophages are often associated with a highly glycolytic metabolism (coupled to extensive lactate secretion as well as NADPH, lipid and nucleotide biosynthesis), and a robust ability to generate reactive oxygen species (ROS), underlying their cytotoxic functions (Andrejeva and Rathmell, 2017). Conversely, M2-like macrophages are generally considered to employ oxidative metabolism for bioenergetic purposes, which has been associated with their ability to support tissue repair (Andrejeva and Rathmell, 2017). However, this oversimplified view does not properly reflect the metabolic heterogeneity of macrophages, nor their functional plasticity in the preservation of tissue homeostasis and immune regulation (Murray, 2017; Murray et al., 2014). Similar considerations apply to TAMs, considerably complicating the delineation of strict metabolic patterns and phenotypic/functional boundaries for M1-like *versus* M2-like TAMs (Cassetta and Pollard, 2018). The metabolic profile of TAMs is indeed very dynamic, and its variations in response to the nutritional needs of malignant cells and TME perturbations have a profound influence not only on TAM survival but also on cancer progression and tumor-targeting immune responses.

Glucose metabolism. TAMs mainly support tumor progression by (1) indirectly increasing the availability of selected nutrients in the TME; (2) providing trophic signals to malignant cells; and (3) mediating robust immunosuppressive functions (**Box 3**). The major mechanism of nutritional support to malignant cells by TAMs is neoangiogenesis, relying on the recruitment or activation of endothelial cells by TAM-derived products including vascular endothelial growth factor A (VEGFA), adrenomedullin (AMD), C-X-C motif chemokine ligand 8 (CXCL8) and CXCL12 (Biswas et al., 2013; Chen et al., 2011; Hughes et al., 2015). Indeed, although the vascular network of developing tumors is

phenotypically and functionally impaired, neoangiogenesis is generally required for neoplasms to grow (Donnem et al., 2018). In this scenario, the TME often displays at least some degree of hypoxia, which promotes the tumor-supporting functions of TAMs via two mechanisms. First, hypoxia favors the upregulation of solute carrier family 40 member 1 (SLC40A1, best known as FPN) and lipocalin 2 (LCN2), as observed both *in vitro* and *in vivo* in human and mouse breast cancer models (Mertens et al., 2016; Mertens et al., 2018; Oren et al., 2016). This results in the acquisition of an iron donor phenotype by TAMs, increased iron availability in the TME, improved iron uptake by malignant cells, and consequent proliferative activation (Mertens et al., 2016; Mertens et al., 2018; Oren et al., 2016). Second, in distinct *in vivo* mouse tumor models, hypoxia drove the upregulation of DNA damage inducible transcript 4 (DDIT4, best known as REDD1), an endogenous inhibitor of mechanistic target of rapamycin (MTOR) complex 1 (MTORC1), in TAMs (Wenes et al., 2016). As a result, hypoxic TAMs shift towards oxidative metabolism coupled to decreased glucose intake, culminating in endothelial cell hyperactivation leading to neoangiogenesis and metastasis as a consequence of increased glucose availability in the TME (Wenes et al., 2016). The physiological relevance of such a shift in glucose metabolism in the human setting is not yet proved.

Even in normoxic conditions, human TAMs display lower glyceraldehyde 3-phosphate dehydrogenase (GAPDH) and succinate dehydrogenase (SDH) activity than normal macrophages, supporting their ability to operate on relatively low nutritional inputs as found in the TME (Miller et al., 2017). Interestingly, GAPDH activity was found to be more pronouncedly decreased in M2-like than in M1-like macrophages infiltrating human colorectal tumors (Miller et al., 2017). Along similar lines, monocyte-derived TAMs from human gliomas exhibited decreased glycolytic metabolism as compared to tissue-resident TAMs, which was associated with increased immunosuppression in the TME and poor patient survival (Muller et al., 2017). These observations suggest that a reduced glycolytic activity in TAMs favors tumor progression via both nutritional and immunological circuitries.

While a reduced flux through glycolysis in TAMs appears to support tumor growth in a variety of settings, co-culture experiments and tissue section analyses demonstrated that lactate production by human medullary thyroid carcinomas cells causes a robust shift from oxidative phosphorylation to glycolysis in TAMs, coupled to increased secretion of lactate, tumor necrosis factor (TNF) and interleukin 6 (IL6), ultimately supporting (rather than inhibiting) disease progression (Arts et al., 2016). Moreover, proteomic analyses revealed that glycolytic enzymes including hexokinase 2 (HXK2) are upregulated both in bone marrow-derived macrophages exposed to breast cancer extracts from patients and in TAMs from individuals with pancreatic cancer, portending to an enhanced (rather than reduced) glycolytic capacity (Liu et al., 2017a; Penny et al., 2016). Such a metabolic reprogramming was linked to increased metastatic dissemination in pancreatic cancer patients (Penny et al., 2016). Thus, glycolysis in TAMs can support tumor growth despite an increased competition for local glucose availability. At least in some murine settings, these observations may reflect the requirement for glycolysis in M2 polarization (Zhao et al., 2017).

Glutamine and fatty acid metabolism. In line with their often poorly glycolytic profile, M2-like TAMs also exhibit elevated glutamine and fatty acid consumption. The former reflects relatively high expression levels of both glutamine transporters and metabolic enzymes, as observed (both *in vitro* and *in vivo*) in mouse tumor models and primary human TAMs (Choi et al., 2015; Colegio et al., 2014). Accordingly, glutamate-ammonia ligase (GLUL) reportedly supports M2 polarization by catalyzing the conversion of glutamate into glutamine, at least *in vitro* (Palmieri et al., 2017). Thus, GLUL inhibition favors the repolarization of M2-like TAMs into their M1-like counterparts accompanied by increased glycolytic flux and succinate availability (Palmieri et al., 2017), suggesting the existence of a metabolic interplay between glucose and glutamine metabolism in the regulation of TAM functions. Moreover, glutamine depletion restrains murine M2 polarization as a consequence of limited α -ketoglutarate availability for epigenetic reprogramming (Jha et al., 2015; Liu et al., 2017b). A similar outcome

ensues the inhibition of *N*-glycosylation, reflecting the limited glucose-, acetyl-CoA- and aspartate-dependent synthesis of UDP-N-acetyl-glucosamine (UDP-GlcNac) (Jha et al., 2015), which also has epigenetic functions (Hardiville and Hart, 2016). The latter results from the interleukin 4 (IL4)-driven activation of signal transducer and activator of transcription 6 (STAT6) and PPARG coactivator 1 beta (PPARGC1B), culminating with increased mitochondrial biogenesis and epigenetic reprogramming toward fatty acid oxidation (FAO) (Vats et al., 2006). Thus, pharmacological FAO inhibition reportedly favors M2-to-M1 repolarization, as shown *in vivo*, in murine models of lung and colorectal cancer (Hossain et al., 2015), while fatty acid synthase (FASN) upregulation in specific murine TAM subsets as a consequence of colony stimulating factor 1 (CSF1) secretion by cancer cells has been shown to support *in vivo* pulmonary tumorigenesis (Park et al., 2015). In this setting, TAMs appear to favor disease progression by releasing the immunosuppressive cytokine interleukin 10 (IL10) downstream of peroxisome proliferator activated receptor delta (PPARD) activation (Park et al., 2015). The latter observation lends further support to the existence of an intimate crosstalk between metabolism and immune functions in the TME.

Apparently at odds with increased FAO utilization, some TAMs (but not cancer cells) accumulate intracellular lipids, supporting not only their metabolic fitness but also their immunomodulatory functions (Xiang et al., 2018). This reflects the deregulation of multiple factors involved in intracellular lipid metabolism, including the enzymes abhydrolase domain containing 5 (ABHD5), monoglyceride lipase (MGLL), and acyl-CoA dehydrogenase medium chain (ACADM, best known as MCAD) (Miao et al., 2016; Niu et al., 2017; Xiang et al., 2018), as well as the chaperones fatty acid binding protein 4 (FABP4) and FABP5 (Hao et al., 2018; Rao et al., 2015). In particular, while TAMs infiltrating early human breast cancers preferentially express FABP5, which is associated with lipid droplet accumulation and secretion of immunostimulatory cytokines including type I interferon (IFN), the macrophage compartment of late-stage tumors exhibits preferential FABP4 expression, which supports

tumor progression by favoring IL6 → STAT3 signaling (Hao et al., 2018). Intriguingly, MCAD inhibition results from the caspase 1 (CASP1)-dependent cleavage of PPARG, at least in some *in vitro* and *in vivo* settings (Niu et al., 2017). As CASP1 is generally activated by immunostimulatory cues (Galluzzi et al., 2016), this latter observation points to the existence of a circuitry whereby immunostimulation in the TME may favor compensatory immunosuppression via a metabolic effect. That said, it remains to be formally established whether TAMs resemble adipocytes in their ability to fuel tumor growth by directly providing lipids to cancer cells (Nieman et al., 2011; Zhang et al., 2018a).

Taken together, these observations delineate the major impact of TAM metabolism on their ability to influence tumor growth, often as a consequence of altered immunomodulation.

Metabolic interactions between TAMs and cancer cells

Cancer cells and TAMs co-exist in the context of a complex, bidirectional metabolic relationship that not only is dictated by, but also impinges on, the immunology of the TME (Cassetta and Pollard, 2018; Coussens et al., 2013; Mantovani et al., 2017; Mazzone et al., 2018)

Signals from cancer cells to TAMs. One of the major cytokines whereby cancer cells condition TAMs to exert immunosuppressive functions is CSF1 (DeNardo et al., 2011). Upon binding to its cognate receptor, CSF1 favors the recruitment of monocyte-derived macrophages to the tumor bed and their polarization toward an M2-like phenotype coupled to (1) FAO upregulation (Park et al., 2015), and (2) secretion of a variety of pro-tumorigenic and immunosuppressive factors, including epidermal growth factor (EGF) (Wyckoff et al., 2004) and IL10 (Ruffell et al., 2014). Accordingly, colony stimulating factor 1 receptor (CSF1R) inhibition with small chemicals or monoclonal antibodies favors the accumulation of M1-like TAMs at the expenses of their M2-like counterparts via multiple mechanisms including direct M2-to-M1 conversion and selective cytotoxicity for M2-like TAMs (although the sensitivity of distinct M2-like subsets is heterogeneous) (Pradel et al., 2018; Pyonteck et al., 2013; Yan et al., 2017). This is accompanied by the restoration of glycolysis, mediates therapeutic effects in a variety of tumor models, and is currently under clinical development for the therapy of solid tumors (DeNardo et al., 2011; Papadopoulos et al., 2017; Ries et al., 2014; Ruffell and Coussens, 2015; Shiao et al., 2015; Strachan et al., 2013). To which extent the beneficial effects of CSF1R inhibitors depend on the metabolic rewiring of TAMs, however, remains to be determined.

The release of CSF1, IL34 and VEGFA (which also condition TAMs to support tumor growth) is particularly sensitive to environmental cues, including chemotherapeutic stress, as well as nutritional metabolic variables, such as nutrient availability, oxygen tension, and local pH (which are largely influenced by vascular dysfunction and lactate secretion) (Colegio et al., 2014; DeNardo et al., 2011;

Henze and Mazzone, 2016). Thus, lactate metabolism is particularly relevant not only for the metabolic symbiosis between hypoxic (lactate generating) and normoxic (lactate importing) cancer cells (Allen et al., 2016), but also for the ability of hypoxic cancer cells to reeducate TAMs toward a poorly glycolytic M2-like profile, exhibiting FAO upregulation, decreased capacity for antigen presentation (Chen et al., 2017a; Colegio et al., 2014; Laoui et al., 2014; Liu et al., 2019) and, at least in glioblastoma, increased expression of immunosuppressive molecules (Kren et al., 2010). Of note, M2 polarization of melanoma-associated TAMs seems to be promoted by a mechanism involving a G protein-coupled receptor (GPCR) that senses TME acidification induced by enhanced cancer cell glycolysis (Bohn et al., 2018). In this content, *in vivo* experiments coupled with mathematical modeling revealed that the ability of TAMs to support neo-angiogenesis largely depends on their distance from existing vessels (Carmona-Fontaine et al., 2017), as this determines their exposure to blood-borne oxygen – which limits VEGFA secretion – *versus* cancer cell-derived factors such as lactate and angiopoietin 2 (ANGPT2) – which promote VEGFA production (Coffelt et al., 2010; Laoui et al., 2014). This spatio-metabolic arrangement has also immunological consequences, as VEGFA favors the expression of immunosuppressive receptors on immune effector cells (see below) (Voron et al., 2015).

Increased lactate availability in the TME (which generally parallels a hypoxic state) also favors the catabolism of arginine by arginase 1 (ARG1) and ARG2 over nitric oxide synthase 2 (NOS2), resulting in increased secretion of tumor-supporting factors (*i.e.*, ornithine, polyamines) by TAMs at the expense of potential anticancer mediators (*i.e.*, nitric oxide, citrulline) (Carmona-Fontaine et al., 2017; Colegio et al., 2014). ARG1 can also be upregulated in M2-like TAMs by danger signals released by apoptotic cancer cells (Galluzzi et al., 2018b), such as sphingosine-1-phosphate (S1P) (Brune et al., 2015), as well as by a FASN-dependent pathway driven by CSF1 (Park et al., 2015). Of note, lactate also favors M2 polarization in mouse breast cancer models by initiating G protein-coupled receptor 132 (GPR132) signaling at the plasma membrane, *i.e.*, operating as a signaling molecule (Chen et al., 2017a).

Accordingly, high levels of GPR132 promote breast cancer infiltration by monocyte-derived macrophages that acquire tumor-supporting functions (Cheng et al., 2016). Yet another lactate receptor, hydroxycarboxylic acid receptor 1 (HCAR1), appears to be upregulated on the surface of highly glycolytic M1-like TAMs (Arts et al., 2016). However, the functional implications of this finding remain obscure.

Signals from TAMs to cancer cells. Importantly, the metabolic influence of cancer cells on TAMs is not unidirectional. Thus, TAMs exposed to hypoxia or lactate secrete multiple cytokines with metabolic functions, including IL6, TNF, C-C motif chemokine ligand 5 (CCL5) and CCL18 (Jeong et al., 2019; Lin et al., 2017; Ye et al., 2018; Zhang et al., 2018c). In particular, while IL6 favors glycolysis by promoting the ability of 3-phosphoinositide dependent protein kinase 1 (PDPK1) to phosphorylate phosphoglycerate kinase 1 (PGK) (Zhang et al., 2018c), TNF, CCL5 and CCL18 boost the synthesis of multiple pro-glycolytic factors including HXK2, PGK1, lactate dehydrogenase A (LDHA), glucose-6-phosphate dehydrogenase (G6PD), pyruvate kinase M1/2 (PKM), pyruvate dehydrogenase kinase 1 (PDK1), pyruvate dehydrogenase (PDH), solute carrier family 2 member 1 (SLC2A1, best known as GLUT1) and vascular cell adhesion molecule 1 (VCAM1) (Jeong et al., 2019; Lin et al., 2017; Ye et al., 2018). Along similar lines, aerobic glycolysis is enhanced, both *in vitro* and *in vivo*, by a mechanism involving transfer of hypoxia inducible factor 1 subunit alpha (HIF1A)-stabilizing long noncoding RNA from lactate-exposed TAMs to neoplastic cells (Chen et al., 2019). Intriguingly, HIF1A appears also to underlie, directly or indirectly, the ability of TAM-derived factors to exacerbate glycolysis in malignant cells (Jeong et al., 2019) as well as the capacity of cancer cell-derived lactate to favor M2 polarization (Colegio et al., 2014). Moreover, M2-like TAMs reportedly contribute to hypoxia in an active manner, reflecting (at least in part) the activation of 5' AMP-activated protein kinase (AMPK) and PPARGC1A and the consequent shift towards a poorly

glycolytic, highly oxidative profile characterized by increased mitochondrial mass and elevated oxygen consumption, as demonstrated in both preclinical and clinical settings (Jeong et al., 2019).

Taken together, these observations delineate an intricate scenario wherein nutritional cues, cancer cells and TAMs mutually influence each other to determine the local organization of the TME (**Figure 1**).

Impact of TAM metabolism on immune responses

The metabolic, functional and immunological landscape of the TME is heterogeneous and evolves over time under the selective pressure of both cell intrinsic and extrinsic factors (Andrejeva and Rathmell, 2017; Biswas, 2015). Two major cell-intrinsic determinants of such a landscape are (1) the precise mutational signature of malignant cells, as specific mutations are known to determine the metabolic profile (Galluzzi et al., 2013); and (2) the activation status of immune cells, as functional changes in multiple immune compartment are associated with (and rely on) major metabolic shifts (Bantug et al., 2018). As discussed above, the impact of TAMs in this scenario is at least dual. Thus, TAMs not only influence the metabolism of cancer cells (both directly and via indirect circuitries), but also impinge on the metabolite pool of the TME for survival.

Immunosuppressive roles of TAMs. Importantly, the metabolic circuitries established between TAMs and cancer cells are instrumental for the maintenance of immunosuppression in the TME (Andrejeva and Rathmell, 2017; O'Neill et al., 2016). Some of the main effectors of anticancer immunity – *i.e.*, helper CD4⁺ T cells, cytotoxic CD8⁺ T cells and natural killer (NK) cells – display a highly glycolytic metabolism on activation, supporting not only energy demands but also the anabolic needs underlying rapid proliferation (Bantug et al., 2018; O'Neill et al., 2016). Conversely, immunosuppressive regulatory T (T_{REG}) cells predominantly rely on oxidative phosphorylation for bioenergetic purposes (Bantug et al., 2018). In this context, cancer cells and immune effector cells compete for limited glucose availability, and M2-like TAMs not only avoid engaging in such competition by preferentially employing oxidative phosphorylation (see above), but also limit glycolytic flux in effector cells by expressing the immunosuppressive molecule CD274 (best known as PD-L1) (Chang et al., 2015; Hartley et al., 2018; Lin et al., 2018; Wagner et al., 2019). PD-L1 is upregulated in TAMs, endothelial cells and malignant cells in response to interferon gamma (IFNG) from effector cells (Chang et al.,

2015; Lane et al., 2018). Moreover, *in vitro* and *in vivo* studies revealed that TAMs can promote PD-L1 expression in mouse and human cancer cells by secreting EGF (Zhang et al., 2017). Interestingly, the interaction of PD-L1 with programmed cell death 1 (PDCD1, best known as PD-1), the co-inhibitory receptor that underlies PD-L1 dependent immunosuppression in T cells, reportedly induces a “retrosignal” that has pro-glycolytic effects on PD-L1⁺ malignant cells (Chang et al., 2015), and limits glycolysis and proliferation in PD-L1⁺ TAMs (Hartley et al., 2018). This delineates a circuitry in which both TAMs and cancer cells react to immune effector functions by tipping the balance of the metabolic competition toward tumor progression. TAMs also express PD-1, and this appears to contribute to the preservation of a tumor-permissive state (Gordon et al., 2017).

PD-L1 expression in TAMs is controlled at the transcriptional level by PKM2 (Palsson-McDermott et al., 2017) and HIF1A (Noman et al., 2014; Palsson-McDermott et al., 2017), as well as by a signal transduction cascade initiated by prostaglandin E₂ (PGE₂) (Prima et al., 2017). PGE₂ is released in the TME as a consequence of increased arachidonic acid metabolism in specific TAM subsets (Daurkin et al., 2011), as well upon the activation of apoptotic mechanisms in cancer cells responding to treatment (Huang et al., 2011). Besides favoring the recruitment of monocyte-derived macrophages to the TME and their polarization toward an M2-like phenotype (at least in some settings) (Heusinkveld et al., 2011; Wen et al., 2015), PGE₂ exerts pro-tumorigenic functions by boosting cancer cell proliferation (Huang et al., 2011; Pennock et al., 2018).

M2-like TAMs also deplete the TME of amino acids for which effector cells are auxotroph, including arginine and tryptophan (Murray, 2016; O'Neill et al., 2016). This reflects the elevated expression of ARG1, ARG2, and the tryptophan-consuming enzyme indoleamine 2,3-dioxygenase 1 (IDO1) by TAMs, and can culminate (at least in some settings) in a robust functional impairment of T and NK cells (Geiger et al., 2016; Labadie et al., 2019). Importantly, the immunomodulatory activity of IDO1

involves not only tryptophan depletion, but also the accumulation of immunosuppressive kynurenine as well as cellular circuitries specifically initiated by IDO1⁺ cells that favor the accumulation of T_{REG} cells (Labadie et al., 2019; Wainwright et al., 2012). TAMs derived from resected human glioblastoma, as well as murine TAMs and immature myeloid cell populations of the TME are also particularly avid of non-essential amino acids such as glutamate, glutamine, serine and cysteine (Choi et al., 2015; Srivastava et al., 2010). These amino acids are required for the optimal effector function of T lymphocytes, meaning that their depletion from the TME subverts immunosurveillance (Choi et al., 2015; Ma et al., 2017; Srivastava et al., 2010; Swamy et al., 2016; Tyrakis et al., 2016).

Lactate secretion also contributes to the establishment of an immunosuppressive TME via a variety of mechanisms including (1) extracellular acidification, resulting in functional anergy and potential demise of T and NK cells (Brand et al., 2016; Harmon et al., 2018); (2) reduced T cell motility as a consequence of impaired glycolytic responses to chemokine receptor signaling (Haas et al., 2015); and (3) polarization of T cells toward immunosuppressive CD4⁺CD25⁺FOXP3⁺ T_{REG} cells, whose prominently oxidative metabolism favors survival in the TME (Angelin et al., 2017; Gerriets et al., 2016). Similar considerations apply to extracellular adenosine. Indeed, M2-like TAMs and other immunosuppressive myeloid and lymphoid cells express high levels of ectonucleoside triphosphate diphosphohydrolase 1 (ENTPD1, best known as CD39) and 5'-nucleotidase ecto (NT5E, best known as CD73), which cooperate to hydrolyze extracellular ATP into adenosine, as well as CD38, which initiates extracellular adenosine synthesis from NAD⁺ (Chevrier et al., 2017; d'Almeida et al., 2016; Karakasheva et al., 2015; Montalban Del Barrio et al., 2016). Adenosinergic signaling mediates a variety of suppressive functions on immune effector cells, hence constituting a promising target for the development of novel therapeutic interventions (Kepp et al., 2017; Vijayan et al., 2017).

An emerging notion is that TAMs also influence non-malignant components of the TME, including CAFs (Hashimoto et al., 2016) and vice versa (Takahashi et al., 2017). Although there is a paucity of data on the metabolic and functional interrelationships between TAMs and stromal cells in the TME, we surmise that such links may impact on disease progression and resistance to therapy. Finally, multiple oncogenes including *MYC* and *KRAS* appear to support tumor progression by driving the recruitment of monocyte-derived macrophages and their polarization toward an M2-like phenotype, as shown in a mouse model of lung tumorigenesis (Kortlever et al., 2017).

Altogether, these observations further corroborate the notion that TAMs and cancer cells co-evolve as they metabolically and immunologically influence each other and various other compartments of the TME (**Figure 2**).

TAM metabolism as a target for cancer immunotherapy

Considerable efforts have been dedicated over the past decade at the development of anticancer immunotherapies based on the depletion/neutralization of M2-like TAMs and/or their repolarization toward an M1-like phenotype (Cassetta and Pollard, 2018; DeNardo and Ruffell, 2019). Such an approach holds great promise, and multiple CSF1R inhibitors are currently under clinical development (**Table 1**). Although these agents were conceived to interrupt CSF1R signaling and the consequent activation of a transcriptional program that support immunosuppression by M2-like TAMs (Coussens et al., 2013; Palucka and Coussens, 2016; Ruffell and Coussens, 2015), inhibition of CSF1R also promotes an extensive metabolic rewiring that culminates with the restoration of glycolysis (Park et al., 2015). Along similar lines, TAM-driven immunosuppression can be relieved, at least partially, by interventions aimed at reducing glycolysis (and hence lactate availability) in the TME, such as the deletion of *LDHA* (Seth et al., 2017) as well as the administration of 2-deoxyglycose (Zhao et al., 2017), MTORC1-targeting agents (Kaneda et al., 2016b) phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit gamma (PIK3CG, best known as PI3K γ) plus PIK3CD (best known as PI3K δ) inhibitors (which jointly cause PKM2 downregulation) (Locatelli et al., 2018). Additional strategies to shift the balance from M2-like to M1-like TAMs include blockade of VEGFA (alone or in combination with ANGPT2), which not only suppresses glycolysis but also inhibits neoangiogenesis in the TME (Kloepper et al., 2016; Peterson et al., 2016; Voron et al., 2015); selective redirection of TAM metabolism toward glycolysis and arginine catabolism via NOS2 (Colegio et al., 2014; Palmieri et al., 2017; Steggerda et al., 2017); pharmacological inhibition of hypoxia (with the hypoxia-activated prodrug TH-302) (Jayaprakash et al., 2018; Popovic et al., 2018); and modulation of iron metabolism (Muliaditan et al., 2018; Zanganeh et al., 2016). Moreover, stimulation of autophagy, which constitutes a valid strategy for stimulating anticancer immune responses (Galluzzi et al., 2017b; Pietrocola et al.,

2017), may favor the differentiation of M1-like TAMs (Esteban-Martinez et al., 2017). That said, the impact of TAM-targeting therapies on the metabolic and immunological profiles of TAMs has not yet fully elucidated. Nonetheless, these examples reinforce the notion that precisely targeted metabolic interventions can reinstate TAM-dependent immunosurveillance.

In this context, targeting is a particularly important issue, for at least two reasons. First, the overall metabolic profile of the TME is dictated by the interplay of malignant cells and multiple non-malignant components (Rybstein et al., 2018), implying that non-targeted agents specific for shared metabolic circuitries, such as glycolysis or FAO, may have net effects that stem from multiple cell compartments (and hence are difficult to anticipate) (Galluzzi et al., 2017c). In line with this notion, MTORC1 inhibitors have been found to paradoxically favor tumor progression as a consequence of glycolysis inhibition in hypoxic TAMs coupled to the activation of a neo-angiogenic program (Wenes et al., 2016). Along similar lines, although the reactivation of glycolysis in M2-like TAMs appears to underlie their repolarization toward an M1-like phenotype, it is tempting to speculate that – given the abundance of TAMs in the TME of most solid tumors – sustained glycolysis activation in this compartment may deprive immune effector cells of glucose, *de facto* mediating immunosuppressive effects (Chang et al., 2015). Second, bioenergetic metabolism displays a high degree of heterogeneity, not only across different, but also within similar TAM subpopulations (Chevrier et al., 2017; Lavin et al., 2017). At least in part, these observations explain the suboptimal efficacy of current TAM-targeting approaches (Cassetta and Pollard, 2018; Quail and Joyce, 2017). Moreover, both innate and acquired resistance mechanisms have been reported, some of which involve compensatory mechanisms connecting to stromal compartments of the TME (Kumar et al., 2017; Quail et al., 2016).

Monoclonal antibodies targeting PD-L1 or PD-1, which are commonly known as immune checkpoint blockers (ICBs), mediate robust immunostimulatory activity and are approved by the US Food and

Drug Administration for use in a variety of oncological indications (Vitale et al., 2019). Although these agents were designed to reverse dysfunction of tumor-infiltrating T cells (Galluzzi et al., 2018a), preclinical and clinical studies indicate that at least some TAM subsets rely on PD-L1 and/or PD-1 (Gordon et al., 2017; Su et al., 2018), and PD-L1 signaling in cancer cells has been reported to stimulate glycolysis (Chang et al., 2015). In this context, the efficacy of PD-L1- and PD-1-targeting ICBs appears to involve not only the reinvigoration of T cell functions, but also (1) the inhibition of glycolysis in malignant cells, resulting in increased glucose availability of TAMs and immune effector cells (Chang et al., 2015), and (2) the inhibition of PD-1 and/or PD-L1 signaling in TAMs, culminating with partial restoration of M1-like functions (Gordon et al., 2017; Hartley et al., 2018). Corroborating the translational relevance of these preclinical findings, the expression levels of PD-L1 on TAMs have been found to influence disease progression in both melanoma and ovarian cancer patients (Lin et al., 2018). Of note, T cells (re)activated by PD-1 targeting ICBs can secrete CSF1 or, at least in the melanoma setting, promote its secretion by malignant cells (Eissler et al., 2016; Neubert et al., 2018), which favors expansion of the M2-like TAM compartment as a determinant to resistance. Alongside, TAM populations expressing high levels of the Fcγ receptor can sequester ICBs to prevent interaction with their pharmacological target, also resulting in resistance (Arlaukas et al., 2017). Taken together, these observations provide a strong rationale for the combined inhibition of CSF1R and PD-1 (DeNardo et al., 2011; Neubert et al., 2018; Strachan et al., 2013; Zhu et al., 2014).

Intriguingly, some human TAM populations appear to have defects in vitamin D metabolism that compromise their ability to mediate oncolytic effects, a deficit that can be compensated for (at least in part) by vitamin D supplementation or administration of the immunomodulatory agent lenalidomide (Bruns et al., 2015; Busch et al., 2018). Finally, a panel of promising therapeutic approaches is being developed to interrupt the immunometabolic crosstalk between malignant cells and TAMs. Such strategies include (but may not be limited to): (1) the concomitant blockage of CCL9 and IL23

(Kortlever et al., 2017) or IL4 and IL13 (DeNardo et al., 2009); (2) the neutralization of TAM-derived CCL5 (Lin et al., 2017); (3) IL6 antagonism (Wang et al., 2018; Zhang et al., 2018c); (4) the downregulation of VCAM1 in malignant cells (Ye et al., 2018); and (5) the abrogation of NF- κ B-dependent immunomodulatory programs in TAMs (Lawrence, 2011). Although in an early stage of development, all these approaches have been shown to support M2-to-M1 macrophage repolarization and hence mediate therapeutic anticancer effects in rodent tumor models.

In summary, TAM metabolism and the immunometabolic circuitries linking TAMs to cancer cells and non-malignant components of the TME stand out as promising therapeutic targets for the development of novel anticancer agents. Based on the observations above, we surmise that the most efficient approaches in this context will simultaneously target several facets of TAM metabolism and immunobiology.

Concluding remarks

It is now clear that metabolic fluctuations in immune cells are intimately connected to their phenotype and function (Bantug et al., 2018; Buck et al., 2017). TAMs are no exception to this rule: they can acquire a broad spectrum of activation states linked to their metabolic profile and exhibit extraordinary plasticity, hence constituting promising targets for the development of novel anticancer therapies (Mantovani et al., 2017; Mazzone et al., 2018; Ruffell et al., 2012).

Current approaches mainly aim at depleting M2-like TAMs and/or favoring their repolarization toward an M1-like phenotype, either as a consequence of direct M2-to-M1 transdifferentiation, or upon the recolonization of the TME by M1-like TAMs in the context of M2-like TAM depletion (Cassetta and Pollard, 2018; Mantovani et al., 2017; Pradel et al., 2018). The efficacy of this approach is limited by the existence of innate and acquired resistance mechanisms, alternative immunosuppressive cells that can compensate for TAMs, and the potential for robust immunosuppression at treatment discontinuation (Bonapace et al., 2014; Quail and Joyce, 2017). As an additional layer of complexity, distinct cell populations of the TME share common metabolic profiles (Bantug et al., 2018; Buck et al., 2017), implying that the sustained modulation of core metabolic pathways may have net immunological effects that are difficult to predict. We surmise that the most effective approaches will have to simultaneously target multiple mechanisms of immunosuppression and - at the same time - provide immunostimulatory cues.

In this context, several metabolic interventions may be harnessed in combination with immunotherapeutic agents to mediate anticancer effects that involve, at least partially, changes in TAM subsets. Promising preclinical results have been obtained with the respiratory complex I inhibitor metformin (Chiang et al., 2017; Ding et al., 2015; Scharping et al., 2017), as well as with modulators of arginine and tryptophan catabolism (Caldwell et al., 2018; Geiger et al., 2016; Ye et al., 2016), vitamin

D and arachidonic acid metabolism (Bruns et al., 2015; Busch et al., 2018), extracellular adenosine and lactate accumulation (d'Almeida et al., 2016; Montalban Del Barrio et al., 2016), and FAO (Niu et al., 2017; Zhang et al., 2018b). Moreover, it appears that M2-like TAMs express high levels of the vacuolar ATPase, a regulator of lysosomal pH that holds promise as a novel drug target (Katara et al., 2014; Kuchuk et al., 2018; Liu et al., 2019).

Several questions on the development of TAM-targeting strategies remain unanswered. First, what are the molecular mechanisms underlying the ability of TAM to rapidly switch their metabolic and functional profile? Second, what is the precise ontogeny of each TAM subpopulation? Third, how does the TAM landscape evolve during disease progression and in response to (immuno)therapy? Finally, what is the best approach to target the metabolic and functional interplays between TAM, immune and stromal cell compartments that support tumor progression?

Future exploration of the temporal and spatial evolution the TAM compartment of tumors that respond to, or progress on, therapy will provide profound insights into the immunometabolism of these cells, potentially elucidating these unknowns and opening hitherto unexplored therapeutic avenues.

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Box 1. Phenotypic and functional plasticity of TAM.

Macrophages exposed to cytokines like IL12, TNF and interferon gamma (IFNG), microbe-associated molecular patterns (MAMPs) such as bacterial lipopolysaccharide (LPS), or other Toll-like receptor (TLR) agonists, acquire a pro-inflammatory (M1) state. Conversely, IL4, IL5, IL10, IL13, CSF1, transforming growth factor beta 1 (TFGB1) and PGE₂ all promote macrophage polarization toward an anti-inflammatory (M2) state. M1 macrophages play a major role in the host defense against infection in the context of T_H1 immunity, while M2 macrophages mainly support T_H2-related tissue repair and remodeling (Murray et al., 2014). Using similar criteria, TAMs can be classified in M1-like (pro-inflammatory and usually anti-tumor) and M2-like (anti-inflammatory and pro-tumor). Emerging evidence indicates that TAMs resemble normal macrophages in their ability to adopt a wide spectrum of intermediate activation states, reflecting the variety of microenvironmental conditions they can be exposed to in the TME (Cassetta and Pollard, 2018; Xue et al., 2014). Moreover, phenotypically and functionally distinct TAM subsets co-existing in the TME often co-express well established M1 and M2 markers (Chevrier et al., 2017; Lavin et al., 2017; Muller et al., 2017). TAMs display a high degree of functional plasticity and can rapidly adapt to microenvironment perturbations as those occurring in the course of tumor progression and (immuno)therapeutic challenges (Andrejeva and Rathmell, 2017; Gubin et al., 2018; Mazzone et al., 2018). Alongside, TAMs can acquire specialized functional states depending on the specific region of the TME they occupy. Thus, while hypoxic and necrotic areas of the TME are enriched for M2-like TAMs with limited capacity for antigen presentation, decreased mobility, and abundant secretion of tumor-supporting factors (Movahedi et al., 2010; Wenes et al., 2016), perivascular areas are characterized by subpopulations of TAMs with robust proangiogenic functions, reflecting high expression levels of TEK receptor tyrosine kinase (TEK, best known as TIE2) (Chen et al., 2017b; Harney et al., 2015; Lewis et al., 2016). Thus, TAMs are characterized by an

extraordinary degree of plasticity that enables them to finely orchestrate their functions in response to microenvironmental cues.

Box 2. Principles of tumor-TAM co-evolution.

The establishment and functional evolution of the TAM compartment are key events for tumor progression. Pro-inflammatory cytokines released in the very early phases of oncogenesis favor the recruitment and polarization of M1-like TAMs, which mediate multipronged anticancer effects (Crusz and Balkwill, 2015; Mantovani and Allavena, 2015). In particular, M1-like TAMs accumulating at sites of early oncogenesis can: (1) produce cytotoxic factors that limit the viability of malignant cells, such as nitric oxide and reactive oxygen species; (2) engulf neoplastic cells and destroy them; and (3) release pro-inflammatory cytokines that further stimulate anticancer immunity (Crusz and Balkwill, 2015; Mantovani and Allavena, 2015). However, a prolonged M1-like TAM activity can foster chronic inflammation, hence promoting genomic instability in malignant cells as a drive to tumor progression (Canli et al., 2017; Elinav et al., 2013; Qian and Pollard, 2010). In this context, cancer cells often acquire the ability to repolarize TAMs toward an M2-like state. Such a “re-education” of the TAM compartment is mediated by mechanisms including (but not limited to): (1) the release of lactate and CSF1 by cancer cells; (2) changes in the T cell contexture; and (3) microenvironmental perturbations (*e.g.*, nutrient deprivation, hypoxia) (Henze and Mazzone, 2016; Mantovani et al., 2017). M2-like TAMs in turn favor tumor progression by secreting growth factors (*e.g.*, EGF), pro-angiogenic molecules (*e.g.*, VEGFA), immunosuppressive factors (*e.g.*, IL10) and proteases that remodel the extracellular microenvironment (*e.g.*, matrix metallopeptidases) (Cassetta and Pollard, 2018; Coussens et al., 2013; Mantovani et al., 2017). High levels of TAMs in the TME are generally associated with high adverse prognosis and/or poor sensitivity to treatment in a variety of solid tumors (Cassetta and Pollard, 2018; DeNardo et al., 2011; Fridman et al., 2017; Mantovani et al., 2017; Ruffell and Coussens, 2015). In specific settings, however, tumor infiltration by M1-like TAMs constitutes a good prognostic factor (Forssell et al., 2007; Quatromoni and Eruslanov, 2012). Similarly, specific TAM

subsets appear to support (or even be required for) the optimal efficacy of various (immuno)therapies (Affara et al., 2014; Asano et al., 2011; Gul et al., 2014; Gunderson et al., 2016; Krieg et al., 2018). Thus, the abundance and functionality of unique TAM subtypes have a major impact on disease outcome in patients affected by a variety of solid neoplasms.

Box 3. Regulators and executors of TAM-dependent immunosuppression.

Regulators. **CSF1R** is tyrosine kinase receptor found on surface of TAMs and other myeloid cell populations (Cannarile et al., 2017). Upon engagement by **CSF1**, CSF1R promotes the recruitment of circulating monocytes, their survival in the TME and their polarization toward an M2-like state (Mantovani et al., 2017). **IL34** is an alternative ligand for CSF1R that resembles CSF1 in its biological functions. **IL4** and **IL13** are T_H2 cytokines abundantly secreted in the TME of established tumors, where they favor the accumulation and maintenance of M2-like TAMs. **PI3K γ** is the prominent isoform of PI3K in leukocytes, and it has been etiologically linked to the transcriptional reprogramming whereby TAMs acquire an immunosuppressive, M2-like state. Such a reprogramming involves the MTORC1-dependent activation of CCAAT enhancer binding protein beta (CEBPB) and integrin subunit alpha 4 (ITGA4) coupled to the inhibition of NF- κ B signaling (De Henau et al., 2016; Foubert et al., 2017; Kaneda et al., 2016a; Kaneda et al., 2016b). LC3-associated phagocytosis (**LAP**) is a specific type of phagocytosis involved in the digestion of unwanted extracellular material (Galluzzi et al., 2017a). The uptake of dying tumor cells by TAMs via LAP contributes to immune evasion by supporting the acquisition of an M2-like state along with the suppression of type I IFN signaling (Cunha et al., 2018).

Executors. M2-polarized TAMs release a variety of anti-inflammatory cytokines (*e.g.*, **IL10** and **TGF β 1**) and chemokines (*e.g.*, **CCL17**, **CCL18** and **CCL22**) that inhibit dendritic cell maturation, hence limiting antigen presentation (Ruffell et al., 2014), and favor the recruitment of immunosuppressive T_{REG} cells (Cassetta and Pollard, 2018). M2-like TAMs also limit immune effector responses through the expression of membrane-bound immunosuppressive molecules such as **PD-L1**, phagocytosis inhibitors such as signal regulatory protein alpha (**SIRPA**), amino acid-catabolizing enzymes (*e.g.*, **ARG1** and **IDO1**), and ecto-enzymes that favor extracellular adenosine accumulation

(*e.g.*, **CD38**, **CD39** and **CD73**). Finally, M2-like TAMs avoid tumor infiltration by cytotoxic T cells (**T cell exclusion**) by mechanisms involving extensive remodeling of the extracellular matrix (often mediated by matrix metallopeptidases) or CD8⁺ T cell trapping (Calderon et al., 2015; Peranzoni et al., 2018; Zhu et al., 2017).

Legends to Figures

Figure 1. Immunometabolic interconnections between cancer cells and tumor-associated macrophages.

As they begin to proliferate uncontrollably, cancer cells consume elevated amounts of glucose for bioenergetic, biosynthetic and antioxidant purposes, which is generally associated with an intense release of lactate in the tumor microenvironment (TME), and acquire the ability to secrete high levels of cancer cell-derived colony stimulating factor 1 (CSF1). CSF1 and lactate favor the repolarization of M1-like tumor-associated macrophages (TAMs) toward an immunosuppressive M2-like state, which is characterized by the release of trophic factors, metabolic modulators and immunosuppressive molecules that foster disease progression, including (but not limited to), epithelial growth factor (EGF), vascular endothelial growth factor A (VEGFA), prostaglandin E₂ (PGE₂), tumor necrosis factor (TNF), C-C motif chemokine ligand 5 (CCL5), CCL18, interleukin 6 (IL6), IL10. Moreover, lactate and glucose deprivation exerts immunosuppressive effects on effector T (T_{EFF}) cells. Alongside, M2-like macrophages upregulate oxidative phosphorylation (OXPHOS) and fatty acid oxidation (FAO) at the expenses of glycolysis, and synthesize abundant levels of arginase 1 (ARG1), ARG2 and indoleamine 2,3-dioxygenase 1 (IDO1). The metabolic shift that accompanies M2 polarization increases glucose availability for cancer cells and aggravates hypoxia, which *vice versa* supports M2 polarization. Ado, adenosine; GZMB, granzyme B; IFNG, interferon gamma; Kyn, kynurenine; NO, nitric oxide; T_{REG}, regulatory T; TNF, tumor necrosis factor.

Figure 2. Immunometabolic co-evolution of cancer cells and tumor-associated macrophages.

Early neoplastic lesions exhibit limited degree of hypoxia, abundant infiltration by effector T cells and a tumor-associated macrophage (TAM) compartment largely polarized toward an immunostimulatory M1-like state. As disease progresses, cancer cells avidly deplete the tumor microenvironment (TME) of glucose as they produce increased amounts of lactate and secrete cytokines that favor the recruitment of

blood-borne monocytes and their polarization toward an immunosuppressive M2-like state. M2-like TAMs exhibit limited phagocytic activity, secrete cytokines and chemokines that support the recruitment of immunosuppressive regulatory T (T_{REG}) cells and neoangiogenesis, deplete the TME of amino acids that are crucial for effector T cells at the same time as they release immunosuppressive molecules such as kynurenine (Kyn) and adenosine (Ado), and favor a remodeling of the extracellular matrix (EMC) that restrains tumor infiltration by effector T cells. Alongside, both cancer cells and M2-like TAMs acquire the ability to express PD-L1, which not only favors T cell exhaustion but also exacerbate metabolic competition in the TME. Ultimately, malignant lesions contain high amounts of M2-like TAMs and immunosuppressive or exhausted T cells, resulting in unrestrained disease progression.

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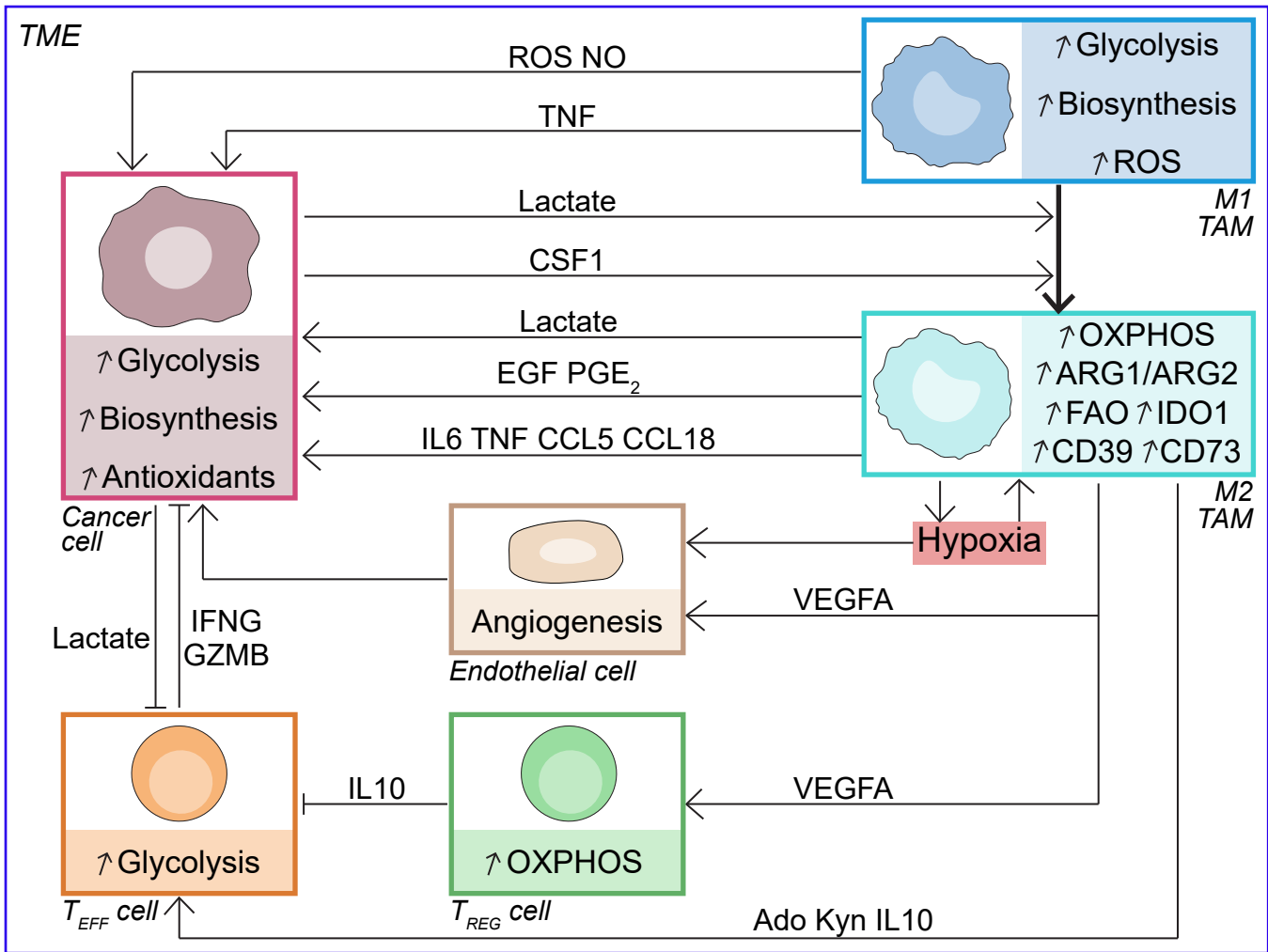


Figure 1

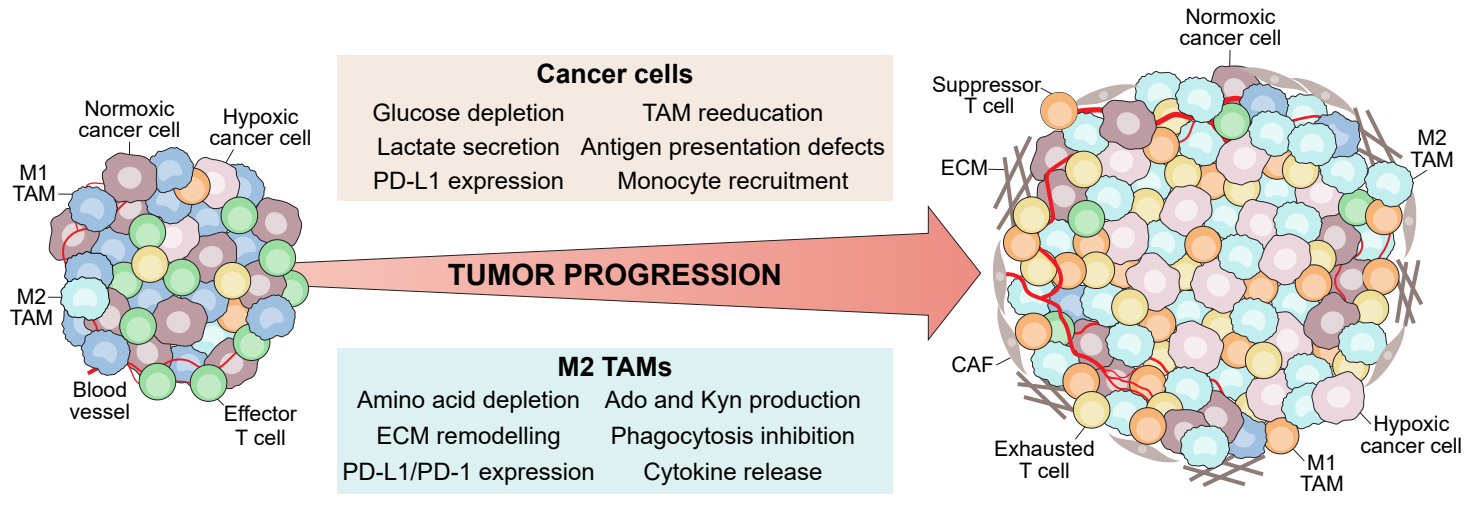


Figure 2

Table 1. CSF1R- and CSF1-targeting agents under clinical development*

Mode of action	Agent	Alias(es)	Number of studies	Progress	Developer
Chemical CSF1R inhibitors	ARRY-382	N/A	Open: 1 Completed: 1 Closed: 0	Phase I/II	Array Biopharma
	BLZ945	N/A	Open: 1 Completed: 0 Closed: 0	Phase I/II	Novartis
	DCC-3014	N/A	Open: 1 Completed: 0 Closed: 0	Phase I	Deciphera Pharmaceuticals
	JNJ-40346527	PRV-6527	Open: 2 Completed: 0 Closed: 0	Phase II	Janssen
	OSI-930	N/A	Open: 0 Completed: 2 Closed: 0	Phase II	Astellas Pharma
	Pexidartinib	PLX3397 PLX108-01	Open: 13 Completed: 6 Closed: 2	Phase III	Plexxikon
	CSF1R-targeting antibodies	AMG820	N/A	Open: 1 Completed: 1 Closed: 0	Phase I/II
Cabiralizumab		FPA008	Open: 10 Completed: 0 Closed: 0	Phase II	FivePrime
Emactuzumab		RO-5509554 RG-7155	Open: 4 Completed: 2 Closed: 1	Phase I/II	Roche
LY3022855		IMC-CS4	Open: 2 Completed: 3 Closed: 0	Phase I/II	Lilly
SNDX-6352		UCB 6352	Open: 1 Completed: 0 Closed: 0	Phase I	Syndax
CSF1-targeting antibodies	Lacnotuzumab	MCS110	Open: 7 Completed: 0 Closed: 2	Phase II	Novartis
	PD 0360324	PD-360,324	Open: 2 Completed: 0 Closed: 0	Phase II	Pfizer

Abbreviations. N/A, not applicable. *as of January 31st, 2019, for oncological indications; source <http://www.clinicaltrials.gov>.