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Linking metabolic reprogramming to therapy resistance in cancer

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Abstract

Metabolic rearrangements are essential to satisfy the different requirements of cancer cells during tumorigenesis and recent studies highlighted a role for such metabolic reprogramming in response and adaptation to therapies. However, therapy-resistant experimental models have been described to be either glycolysis-dependent or OXPHOS-addicted. Here we discuss the recent literature on metabolic reprogramming of cancer in therapy resistance with a plausible explanation of the observed differences which collectively indicate that dis-regulated metabolic pathways could be considered as potential therapeutic target in tumors resistant to conventional therapy.

Introduction

Preventing or bypassing drug resistance is arguably the most important unmet medical need in cancer management. Targeted therapies have been used effectively both as monotherapy and/or in combination with chemotherapy in the vast majority of tumors. However, resistance still limits their clinical benefit and patients experience relapse that includes distant metastases caused by resistant clones unaffected by the selective pressure of the therapy and able to repopulate the tumor. Identification of biomarker-defined patient populations that will most likely respond to the drugs is therefore essential (i) to maximize benefit from targeted therapies and (ii) to minimize exposure of patients to unnecessary treatments that have important side-effects and are expensive.

Metabolic deregulation is an established hallmark of cancer. It is well documented that most cancer cells enhance glucose and glutamine consumption to satisfy their energy demand and biosynthesis requirements for rapid proliferation. Importantly, Warburg reported that, even in the presence of oxygen, cancer cells show increased glycolysis using only a small fraction of glucose for oxidative phosphorylation (OXPHOS). However, whether such metabolic deregulation towards a Warburg-like metabolism is a requisite of the population of cancer cells responsible for therapy adaptation, residual disease and ultimately tumor relapse, remains a matter of debate.

Importantly, most studies that link metabolism and therapy resistance are correlative and did not directly prove that metabolic reprogramming is causative and not a mere bystander effect of signaling and proliferative inputs that characterize resistant cancer cells. On the other hand, resistant cell subpopulations in different types of tumors have been described to be either Warburg-like or OXPHOS-addicted, with plausible explanations as to which metabolic phenotype could be advantageous during tumor development and adaptation to therapy. The aim of the current perspective is to discuss the emerging literature on

metabolic reprogramming in therapy response and resistance and to give a tentative answer to such *conundrum*.

We reviewed recent studies on this topic, which collectively suggest that metabolic reprogramming that resistant cancer cells undergo is profoundly influenced by the type of therapy. Crucially, we speculate that therapy could have an active role in selecting resistant clones and, depending on the mechanisms of action, drugs could confer to the cancer cells a high degree of plasticity making them extremely skillful in rewiring their metabolic network. For the sake of completeness, it is well established that cancer cells can catabolize nutrients other than glucose for energy production and anabolic purposes. However, the current perspective focuses exclusively on central carbon metabolism in therapy resistance.

Chemotherapy resistance

Chemotherapy acts by interfering with cancer cells proliferation at different phases of cell division. Since cell proliferation occurs also in certain normal tissues, side effects during chemotherapy are common. Several chemotherapy resistance mechanisms have been described and can generally be divided into genetic, i.e. gene mutation or amplification that renders the drugs ineffective on a particular subpopulation of cancer cells [1], and non-genetic, in which cancer cells may find a way to bypass the blockade induced by the drug and/or decrease the amount of drug inside the cell by reducing its intracellular transport or by pumping the drug out via multidrug resistance protein transporters [2]. From a metabolic point of view, it is established that a highly proliferative tumor relies on aerobic glycolysis to sustain a fast growth rate [3]. This is one of the reasons why chemotherapy becomes selective for cancer cells. Many studies, initially performed in established cancer cell lines, showed that resistant cells are characterized by aerobic glycolysis and that lactate levels, as by-products of glycolysis, are enhanced in drug-

resistant or metastatic cancers, which implies that the Warburg effect in these cancers may reflect metabolic adaptations associated with development of resistance to chemotherapy [4-6]. Nevertheless, due to the toxic effects on highly proliferating cells exerted by chemotherapy, it is conceivable that a cancer cell has to reduce its proliferation speed and switch to OXPHOS metabolism, which is characterized by a significantly lower glucose demand rate but constitutes a more efficient source for energy generation. Therefore, the emerging idea is that chemotherapy induces a selective pressure, promoting emergence of a subpopulation of cancer cells capable of surviving in the presence of the drug, possibly by efficiently generating the ATP necessary for pumping the drug out of the cell, slowing down the cell cycle with an increase in the population at the G0 phase in order to avoid cell death. Indeed, several recent studies have shown that chemotherapy-resistant cancer cells become OXPHOS-dependent [7-11] . Additionally, increased ATP levels and lower proliferation rate of cancer cells are positively correlated with chemo-resistance [12].

Several studies pointed out that cancer stem cells (CSC) are responsible for therapy resistance and tumor relapse. Understanding CSC metabolism could therefore offer the possibility to target such aggressive subpopulations by interfering with their metabolic features. As recently reviewed by Sancho and co-workers [13], contradictory results described the CSC metabolic phenotype as glycolytic or OXPHOS-addicted not only in various tumor types, but also within individual cancer types. In any case, it is conceivable that OXPHOS dependency could confer a selective advantage to CSCs in the context of specific tumor microenvironments. Indeed, CSCs could uptake lactate and other carbon sources secreted by more differentiated cancer cells or by stromal cells that preferentially undergoes Warburg-like metabolism [14-16]. Despite the fact that the theoretical rationale of this hypothesis seems to be flawless, most of the data provided on chemotherapy-

resistant specimens revealed an increase in glycolytic-related markers in the resistant patients' cohort [6, 17-19]. A possible explanation could be that few persistent cancer cells that survived initial drug selection and are undetectable by current diagnostic approaches, may have already switched to a fast-growing mode once clinical relapse is detectable, and glycolytic transit amplifying cancer cells may actually account for the majority of the tumor bulk at this stage (Figure 1).

Endocrine therapy resistance

Endocrine therapy is the standard of care for breast cancer in which estrogen receptor (ER) is expressed and controls cancer growth and survival. Such therapy acts by interfering with ER signaling either by antagonizing binding of E2 to the ER (e.g., tamoxifen), promoting ER degradation (e.g., fulvestrant), or blocking E2 biosynthesis (aromatase inhibitors, AI). However, resistance limits its clinical efficacy. The mechanisms of such resistance differ substantially to that described for chemotherapeutic agents since the role of hormones in adult tissue, despite being fundamental, impacts only on certain tissues. Although genetic events have been reported to concur to therapy resistance [20, 21], the majority of the resistant events are caused by activation of ER signaling independently of estrogen binding [22]. Such signaling rewiring is paralleled by metabolic reprogramming of the resistant cells. It has been shown that tamoxifen-resistant cells are characterized by Hypoxia-Inducible Factor-1α (HIF-1α) hyper-activation via modulation of Akt/mTOR, resulting in enhanced aerobic glycolysis and a Warburg-like metabolism. Importantly, impairing glycolysis restores tamoxifen sensitivity in drug-resistant cells, suggesting that metabolic reprogramming is not merely a consequence of signaling rewiring [23]. Moreover, a different study reported that when tamoxifen-sensitive cells are cultured in the presence of fibroblasts, a metabolic symbiosis is established, impacting on drug response. Indeed, it has been proposed that while cancer associated fibroblasts are

undergoing a Warburg-like metabolism, secreting lactate into the media, cancer cells are able to uptake such carbon sources, thus switching to OXPHOS, and this is sufficient to confer tamoxifen resistance [24]. However, such interactions also generate a growth factors/cytokines crosstalk between the cell populations which is difficult to ignore when analyzing the resistance mechanisms and makes it difficult to draw a definitive conclusion on metabolic reprogramming [25]. What seems plausible in endocrine therapy resistance comes from recent studies where metabolic reprogramming of resistant cells shows high degree of plasticity. Accordingly to Bacci and co-workers, although AI resistant cells undergo a typical Warburg-like metabolism characterized by enhanced basal glucose uptake, glycolysis targeting in such cells is ineffective [18]. Indeed, resistant cells were able to shift *ad hoc* between Warburg and OXPHOS metabolism, a phenomenon directed by the microRNA-155 (Figure 2). Glycolytic parameters were found to correlate with endocrine therapy response and microRNA-155 levels display prognostic value [18]. Sansone and co-workers' study has further supported this "chameleonic" metabolic behavior of endocrine therapy resistant cells [26]. Crucially, they reported that endocrine therapy selected a metabolic dormant cancer stem cell-like subpopulation characterized by the loss of mitochondrial bioenergetics. Importantly, the exit from this metabolic dormancy is orchestrated by IL6 signaling that impacts on ER expression and function and culminates in reacquisition of glycolytic and OXPHOS metabolic activity. Taken together, these data highlight the importance of metabolic adaptability of cancer cells for endocrine therapy resistance and suggest that targeting such metabolic plasticity could be a novel approach to combat and/or delay disease recurrence.

Antiangiogenic therapy resistance

It is established that administration of antiangiogenic therapy causes a rise in hypoxia in the targeted tissues and subsequent HIF1α-mediated upregulation of glycolysis genes,

including glucose transporter 1 (GLUT1), lactate dehydrogenase A (LDHA), monocarboxylate transporter 4 (MCT4) [27, 28]. More recent data further support the hypothesis that antiangiogenic drugs might rewire tumor metabolism, including both studies which exploited vascular endothelial growth factor (VEGF) neutralizing antibodies, such as bevacizumab [29], and those involving antiangiogenic tyrosine kinase inhibitors (TKIs), such as sunitinib or nintedanib, which target multiple receptors involved in angiogenesis [30-32]. As mentioned above, hypoxic tumor areas are characterized by increased expression of HIF1α and its target MCT4, a lactate transporter which is often over-expressed by cells surrounding necrotic areas in solid cancers. In contrast, tumor areas relatively distant from the hypoxic zone preferentially express MCT1 - which predominantly acts as monocarboxylate importer [33] - and behave as OXPHOS-dependent regions. This pattern of tissue expression of MCT isoforms reflects a metabolic mosaicism, also termed metabolic symbiosis [30-32], which could derive from the effects of the tumor microenvironment, in particular the availability of oxygen and nutrients, on the metabolic plasticity of tumor cells (Figure 1).

Notably, however, in some experimental tumors treated with antiangiogenic drugs, such as bevacizumab, there is evidence of the existence of $MCT4^{+}$ tumor regions uncoupled from necrotic and *bona fide* hypoxic areas [36]. This observation, along with the metabolic characterization of *ex vivo* tumor cells obtained from tumors treated with anti-VEGF therapy [29], raises the hypothesis that in parallel with hypoxia-driven metabolic plasticity, antiangiogenic therapy could elicit *in vivo* selection of tumor cells with stable metabolic changes compared with the pre-treatment tumor, such as enhanced glycolysis [34]. This hypothesis underscores the possible existence of intra-tumor metabolic heterogeneity not only imposed by local pathophysiological conditions, such as hypoxia or glucose deprivation, or adaptation to a specific environment [35], but rather intrinsic to tumor cells. This type of metabolic heterogeneity is still poorly characterized and it could be connected

with the genetic heterogeneity of solid tumors, given the established link between certain oncogenes/tumor suppressors and metabolism (such as Akt/KRAS/glycolysis and MYC/glutaminolysis), or, alternatively, be accounted for by epigenetic mechanisms [36].

One shared conclusion of the above quoted studies is the association between these metabolic changes and resistance to angiogenesis inhibition [37]. This novel mechanism of resistance is substantially new and it is not due to the bypassing of the vasculature blockade, given the fact that in the various models tumor growth occurred despite persistently reduced vascularization.

Intriguingly, genetic targeting of either the glycolytic part of the tumor (i.e. by genetic inactivation of MCT4) or the OXPHOS tumor region (by mTOR activity blockade) improved therapeutic activity of angiogenesis inhibitors. These results corroborate previous studies, which showed the possibility of improving therapeutic activity of sorafenib by modulating glycolysis with dichloroacetate (DCA) in hepatocellular carcinoma models [38] or by administration of bevacizumab plus drugs targeting carbonic anhydrase IX, an enzyme involved in pH regulation, in colon and glioblastoma models [39]. Along this line, some immunomodulatory drugs such as thalidomide and its derivatives have been recently shown to destabilize the cereblon-CD47-MCT1 axis [40], raising the possibility of exploiting thalidomide or related drugs to counteract increased MCT1/MCT4 expression associated with antiangiogenic therapy. Additional therapeutic approaches could focus on glycolysis-targeting drugs, such as LDHA inhibitors [41], which could possibly disclose enhanced therapeutic activity in tumors pre-treated with angiogenesis inhibitors, compared to untreated tumors, although some safety concerns exist related to these drugs and the field suffers from an historical lack of interest by the pharmaceutical industry [42].

Finally, since acquisition of this detrimental glycolytic phenotype seems to require chronic administration of the antiangiogenic drug, one may ask whether it might be possible to delay it by changing the schedule of administration of these drugs. Although interruption of

antiangiogenic therapy has been shown to negatively impact on angiogenesis control [43], from the viewpoint of metabolism it will be important to investigate how an intermittent schedule of treatment impacts on hypoxia, HIF1-α accumulation and metabolic mosaicism. One limitation of the published studies is that they were generally not designed to detect additional metabolic perturbations caused by antiangiogenic therapy, not involving glycolysis. In one study, however, Hanahan *et a*l. investigated the metabolic profile of sunitinib-treated versus control tumors by a metabolomic approach, but they failed to disclose clear differences, assigning this negative finding to the intra-tumor metabolic heterogenicity [30]. Others, such as Keunen *et al*., investigated by magnetic resonance spectroscopy the metabolic profile of glioblastoma xenografts treated with bevacizumab and found an increase in lactate and alanine metabolites, together with HIF-1α induction and PI3K/AKT pathway activation [44]. Finally, Sounni *et al*. investigated the metabolic perturbations associated with sorafenib in breast cancer models and disclosed increased lipid metabolism during the re-oxygenation phase [45], whereas Bensaad *et al*. described up-regulation of fatty acid uptake and metabolism in tumors treated with bevacizumab, involving increased expression of FATBP3 and FATBP7 and accumulation of lipid droplets in tumor cells [46]. Given the existence of metabolic heterogeneity, which could represent a confounding factor, future studies in this area would greatly profit of new imaging methods, such as certain NMR techniques, which enable topographic imaging of metabolism in tumors [47]. In any case, whether these additional metabolic alterations contribute to acquired resistance to antiangiogenic drugs is currently unknown.

Targeted therapy and metabolism changes

Apart from antiangiogenic therapy, also resistance to other targeted therapies seems to involve metabolic adaptation of tumors cells. For instance, resistance of breast cancer cells to the HER-2-targeting trastuzumab has been associated with increased glycolysis

via heat shock factor 1 and LDHA up-regulation and combining trastuzumab with glycolysis inhibitors synergistically inhibited trastuzumab-sensitive and -resistant breast cancers both *in vitro* and *in vivo* [48].

Along this line, pancreatic adenocarcinoma cell lines resistant to the multi-tyrosine kinase inhibitor axitinib show increased activated Akt and up-regulation of glucose uptake *in vitro*, due to the membrane re-localization of the glucose transporter GLUT1, as well as increased glycolytic activity [49]. Indeed, interference with a phosphatidylinositol-3 kinase (PI3K) inhibitor reversed the GLUT1 translocation and restored sensitivity to axitinib treatment.

Finally, a recent study on NOTCH1-addicted T acute lymphoblastic leukemia (T-ALL) disclosed that resistance to γ-secretase inhibitors (GSI) - a class of drugs able to block NOTCH1 signaling - was associated with prominent metabolic reprogramming involving both glycolysis and glutaminolysis in a mouse model of Pten-deficient T-ALL [50]. Moreover, genetic and pharmacologic inhibition of glutaminase was highly synergistic with inhibition of NOTCH1 signaling in T-ALL cell lines and patient-derived T-ALL xenografts. More broadly, these results underscore the importance of metabolic rewiring in the context of resistance to targeted therapies and demonstrate that secondary mutations, such as those in PTEN in the case of T-ALL, can greatly impact of the metabolic profile of tumors. It can be speculated that ongoing studies will soon uncover similar mechanisms in other types of tumors resistant to targeted therapy.

Conclusions

Although it is widely accepted that metabolic rearrangements are essential to satisfy the different requirements of a cancer cell during tumorigenesis, the metabolic reprogramming in response and adaptation to anti-tumor therapies has been perceived as a bystander effects of molecular and genetic rearrangements. The current manuscript gathers recent

findings in metabolic reprogramming and anti-tumor therapy response and suggests that such reprogramming is essential for the acquisition of a resistant phenotype. Importantly, different drugs can select different metabolic phenotypes in the resistant cancer cells ultimately impacting on tumor plasticity: such plasticity seems to be crucial to resist the stress induced by therapies. In summary, it is plausible to affirm that the ability to rewire proliferative and pro-survival networks that characterizes aggressive/resistant cancer cells is functional to confer them a high degree of metabolic plasticity to respond and react to external stress stimuli, such as those triggered by anti-tumor agents.

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References

1. Sakai, W., et al., *Secondary mutations as a mechanism of cisplatin resistance in BRCA2-mutated cancers.* Nature, 2008. **451**(7182): p. 1116-20.
2. Ozben, T., *Mechanisms and strategies to overcome multiple drug resistance in cancer.* FEBS Lett, 2006. **580**(12): p. 2903-9.
3. Vander Heiden, M.G., L.C. Cantley, and C.B. Thompson, *Understanding the Warburg effect: the metabolic requirements of cell proliferation.* Science, 2009. **324**(5930): p. 1029-33.
4. Xu, R.H., et al., *Inhibition of glycolysis in cancer cells: a novel strategy to overcome drug resistance associated with mitochondrial respiratory defect and hypoxia.* Cancer Res, 2005. **65**(2): p. 613-21.

5. Wagner, W., W.M. Ciszewski, and K.D. Kania, *L- and D-lactate enhance DNA repair and modulate the resistance of cervical carcinoma cells to anticancer drugs via histone deacetylase inhibition and hydroxycarboxylic acid receptor 1 activation.* Cell Commun Signal, 2015. **13**: p. 36.
6. Botzer, L.E., et al., *Hexokinase 2 is a determinant of neuroblastoma metastasis.* Br J Cancer, 2016. **114**(7): p. 759-66.
7. Giannoni, E., et al., *Targeting stromal-induced pyruvate kinase M2 nuclear translocation impairs oxphos and prostate cancer metastatic spread.* Oncotarget, 2015. **6**(27): p. 24061-74.
8. Viale, A., et al., *Oncogene ablation-resistant pancreatic cancer cells depend on mitochondrial function.* Nature, 2014. **514**(7524): p. 628-32.
9. Matassa, D.S., et al., *Oxidative metabolism drives inflammation-induced platinum resistance in human ovarian cancer.* Cell Death Differ, 2016. **23**(9): p. 1542-54.
10. Vellinga, T.T., et al., *SIRT1/PGC1α-Dependent Increase in Oxidative Phosphorylation Supports Chemotherapy Resistance of Colon Cancer.* Clin Cancer Res, 2015. **21**(12): p. 2870-9.
11. Ippolito, L., et al., *Metabolic shift toward oxidative phosphorylation in docetaxel resistant prostate cancer cells.* Oncotarget, 2016.
12. Zhou, Y., et al., *Intracellular ATP levels are a pivotal determinant of chemoresistance in colon cancer cells.* Cancer Res, 2012. **72**(1): p. 304-14.
13. Sancho, P., D. Barneda, and C. Heeschen, *Hallmarks of cancer stem cell metabolism.* Br J Cancer, 2016. **114**(12): p. 1305-12.
14. Martinez-Outschoorn, U.E., M.P. Lisanti, and F. Sotgia, *Catabolic cancer-associated fibroblasts transfer energy and biomass to anabolic cancer cells, fueling tumor growth.* Semin Cancer Biol, 2014. **25**: p. 47-60.

15. Fiaschi, T., et al., *Reciprocal metabolic reprogramming through lactate shuttle coordinately influences tumor-stroma interplay.* Cancer Res, 2012. **72**(19): p. 5130-40.
16. Fong, M.Y., et al., *Breast-cancer-secreted miR-122 reprograms glucose metabolism in premetastatic niche to promote metastasis.* Nat Cell Biol, 2015. **17**(2): p. 183-94.
17. Bovenzi, C.D., et al., *Prognostic Indications of Elevated MCT4 and CD147 across Cancer Types: A Meta-Analysis.* Biomed Res Int, 2015. **2015**: p. 242437.
18. Bacci, M., et al., *miR-155 drives metabolic reprogramming of ER+ breast cancer cells following long-term estrogen deprivation and predicts clinical response to aromatase inhibitors.* Cancer Res, 2016.
19. Doyen, J., et al., *Expression of the hypoxia-inducible monocarboxylate transporter MCT4 is increased in triple negative breast cancer and correlates independently with clinical outcome.* Biochem Biophys Res Commun, 2014. **451**(1): p. 54-61.
20. Fribbens, C., et al., *Plasma ESR1 Mutations and the Treatment of Estrogen Receptor-Positive Advanced Breast Cancer.* J Clin Oncol, 2016. **34**(25): p. 2961-8.
21. Alluri, P.G., C. Speers, and A.M. Chinnaiyan, *Estrogen receptor mutations and their role in breast cancer progression.* Breast Cancer Res, 2014. **16**(6): p. 494.
22. Morandi, A., et al., *GDNF-RET signaling in ER-positive breast cancers is a key determinant of response and resistance to aromatase inhibitors.* Cancer Res, 2013. **73**(12): p. 3783-95.
23. Woo, Y.M., et al., *Inhibition of Aerobic Glycolysis Represses Akt/mTOR/HIF-1α Axis and Restores Tamoxifen Sensitivity in Antiestrogen-Resistant Breast Cancer Cells.* PLoS One, 2015. **10**(7): p. e0132285.
24. Martinez-Outschoorn, U.E., et al., *Understanding the metabolic basis of drug resistance: therapeutic induction of the Warburg effect kills cancer cells.* Cell Cycle, 2011. **10**(15): p. 2521-8.

25. Morandi, A. and P. Chiarugi, *Metabolic implication of tumor:stroma crosstalk in breast cancer.* J Mol Med (Berl), 2014. **92**(2): p. 117-26.
26. Sansone, P., et al., *Self-renewal of CD133(hi) cells by IL6/Notch3 signalling regulates endocrine resistance in metastatic breast cancer.* Nat Commun, 2016. **7**: p. 10442.
27. Bergers, G. and D. Hanahan, *Modes of resistance to anti-angiogenic therapy.* Nat Rev Cancer, 2008. **8**(8): p. 592-603.
28. McIntyre, A. and A.L. Harris, *Metabolic and hypoxic adaptation to anti-angiogenic therapy: a target for induced essentiality.* EMBO Mol Med, 2015. **7**(4): p. 368-79.
29. Curtarello, M., et al., *VEGF-targeted therapy stably modulates the glycolytic phenotype of tumor cells.* Cancer Res, 2015. **75**(1): p. 120-33.
30. Allen, E., et al., *Metabolic Symbiosis Enables Adaptive Resistance to Anti-angiogenic Therapy that Is Dependent on mTOR Signaling.* Cell Rep, 2016. **15**(6): p. 1144-60.
31. Jimenez-Valerio, G., et al., *Resistance to Antiangiogenic Therapies by Metabolic Symbiosis in Renal Cell Carcinoma PDX Models and Patients.* Cell Rep, 2016. **15**(6): p. 1134-43.
32. Pisarsky, L., et al., *Targeting Metabolic Symbiosis to Overcome Resistance to Anti-angiogenic Therapy.* Cell Rep, 2016. **15**(6): p. 1161-74.
33. Perez-Escuredo, J., et al., *Lactate promotes glutamine uptake and metabolism in oxidative cancer cells.* Cell Cycle, 2016. **15**(1): p. 72-83.
34. Quintieri, L., M. Selmy, and S. Indraccolo, *Metabolic effects of antiangiogenic drugs in tumors: therapeutic implications.* Biochem Pharmacol, 2014. **89**(2): p. 162-70.
35. Davidson, S.M., et al., *Environment Impacts the Metabolic Dependencies of Ras-Driven Non-Small Cell Lung Cancer.* Cell Metab, 2016. **23**(3): p. 517-28.

36. Pavlova, N.N. and C.B. Thompson, *The Emerging Hallmarks of Cancer Metabolism.* Cell Metab, 2016. **23**(1): p. 27-47.
37. Rapisarda, A. and G. Melillo, *Role of the hypoxic tumor microenvironment in the resistance to anti-angiogenic therapies.* Drug Resist Updat, 2009. **12**(3): p. 74-80.
38. Shen, Y.C., et al., *Activating oxidative phosphorylation by a pyruvate dehydrogenase kinase inhibitor overcomes sorafenib resistance of hepatocellular carcinoma.* Br J Cancer, 2013. **108**(1): p. 72-81.
39. McIntyre, A., et al., *Carbonic anhydrase IX promotes tumor growth and necrosis in vivo and inhibition enhances anti-VEGF therapy.* Clin Cancer Res, 2012. **18**(11): p. 3100-11.
40. Eichner, R., et al., *Immunomodulatory drugs disrupt the cereblon-CD147-MCT1 axis to exert antitumor activity and teratogenicity.* Nat Med, 2016. **22**(7): p. 735-43.
41. Granchi, C. and F. Minutolo, *Anticancer agents that counteract tumor glycolysis.* ChemMedChem, 2012. **7**(8): p. 1318-50.
42. Martinez-Outschoorn, U.E., et al., *Cancer metabolism: a therapeutic perspective.* Nat Rev Clin Oncol, 2016.
43. Mancuso, M.R., et al., *Rapid vascular regrowth in tumors after reversal of VEGF inhibition.* J Clin Invest, 2006. **116**(10): p. 2610-21.
44. Keunen, O., et al., *Anti-VEGF treatment reduces blood supply and increases tumor cell invasion in glioblastoma.* Proc Natl Acad Sci U S A, 2011. **108**(9): p. 3749-54.
45. Sounni, N.E., et al., *Blocking lipid synthesis overcomes tumor regrowth and metastasis after antiangiogenic therapy withdrawal.* Cell Metab, 2014. **20**(2): p. 280-94.
46. Bensaad, K., et al., *Fatty acid uptake and lipid storage induced by HIF-1alpha contribute to cell growth and survival after hypoxia-reoxygenation.* Cell Rep, 2014. **9**(1): p. 349-65.

47. Sengupta, D. and G. Pratx, *Imaging metabolic heterogeneity in cancer.* Mol Cancer, 2016. **15**: p. 4.

48. Zhao, Y., et al., *Overcoming trastuzumab resistance in breast cancer by targeting dysregulated glucose metabolism.* Cancer Res, 2011. **71**(13): p. 4585-97.

49. Hudson, C.D., et al., *Resistance to the tyrosine kinase inhibitor axitinib is associated with increased glucose metabolism in pancreatic adenocarcinoma.* Cell Death Dis, 2014. **5**: p. e1160.

50. Herranz, D., et al., *Metabolic reprogramming induces resistance to anti-NOTCH1 therapies in T cell acute lymphoblastic leukemia.* Nat Med, 2015. **21**(10): p. 1182-9.

Figure 1

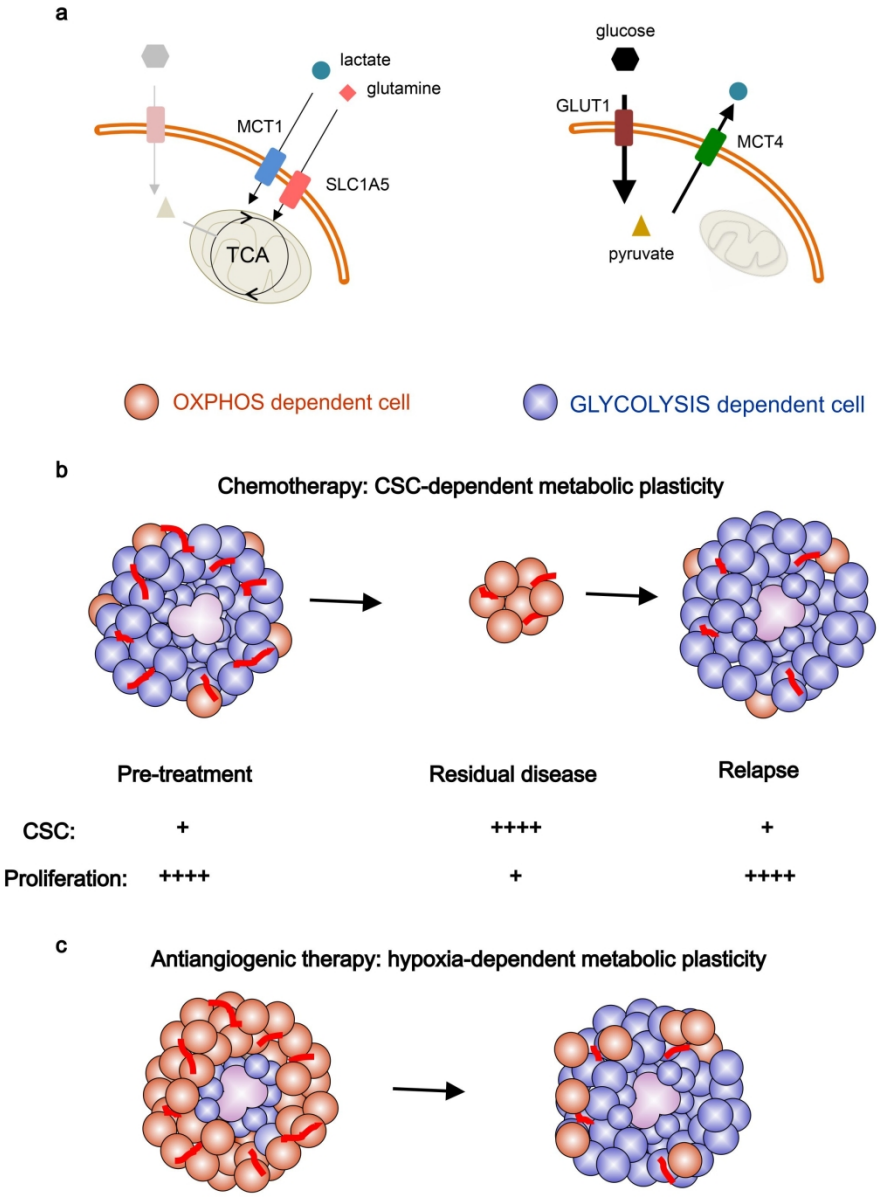

Figure 1. Metabolic plasticity in the context of cancer therapy. Before treatment, it is envisioned that both OXPHOS- and glycolysis-dependent cells co-exist in the tumor (a). Chemotherapy may impact on cancer stem cell (CSC)-dependent metabolic plasticity. According to this model, the final metabolic phenotype of the tumor depends on its content of CSC versus non-CSC populations (b). In contrast, antiangiogenic therapy could primarily trigger hypoxia-dependent metabolic plasticity, involving topographic distribution of cancer cells endowed with different metabolic phenotype in tumor areas exposed to various concentrations of oxygen and nutrients (c).

Figure 2

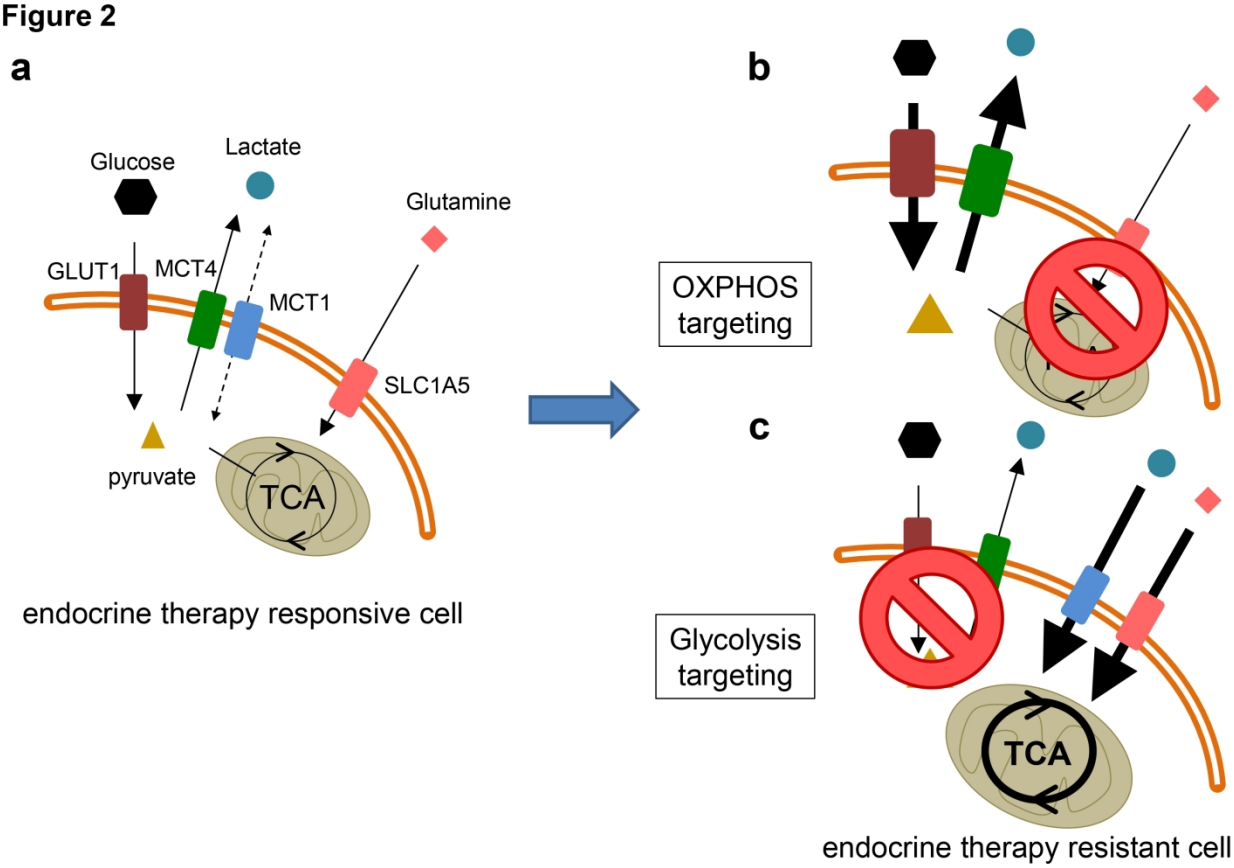

Figure 2. ER positive endocrine therapy (ET) responsive breast cancer cells are characterized by a basal metabolism that predominantly undergoes OXPHOS with a partial release of lactate (a). Indeed, these cells are sensitive to both glycolysis and OXPHOS blockade. When becoming ET-resistant, cancer cells display higher uptake of glucose and are insensitive to OXPHOS targeting (b). However, targeting glycolysis is substantially ineffective, due to the ability of cancer cells to rewire their metabolic pathways, thus acquiring OXPHOS dependency (c).