Published in final edited form as:

Am Soc Clin Oncol Educ Book. 2017 ; 37: 825–832. doi:10.14694/EDBK_175561.

Metabolic Alterations In Cancer and Their Potential As Therapeutic Targets

Jamie D. Weyandt, PhD¹, Craig B. Thompson, MD², Amato J. Giaccia, PhD³, and W. Kimryn Rathmell, MD, PhD¹

¹Department of Medicine, Division of Hematology and Oncology, Vanderbilt-Ingram Cancer Center, Vanderbilt University Medical Center, Nashville, TN 37232

²Cancer Biology and Genetics Program, Memorial Sloan-Kettering Cancer Center, New York, New York, 10065

³Department of Radiation Oncology, Stanford University School of Medicine, Stanford, CA 94305

Abstract

Otto Warburg's discovery in the 1920's that tumor cells took up more glucose and produced more lactate than normal cells provided the first clues that cancer cells reprogrammed their metabolism. For many years, however, it was unclear as to whether these metabolic alterations were a consequence of tumor growth, or an adaptation that provided a survival advantage to these cells. In more recent years, interest in the metabolic differences in cancer cells has surged, as tumor proliferation and survival has been shown to be dependent upon these metabolic changes. In this educational review, we discuss some of the mechanisms that tumor cells use for reprogramming their metabolism to provide the energy and nutrients that they need for quick or sustained proliferation, and discuss the potential for therapeutic targeting of these pathways to improve patient outcomes.

Keywords

Cancer; metabolism; mTOR; hypoxia; glycolysis; IDH

Metabolism in Normal vs. Tumor Cells: The Warburg Effect and Beyond

Metabolic pathways are the means by which cells break down nutrients to acquire the energy and building blocks that they need for growth, proliferation, and the maintenance of critical cellular processes. Energy within cells is stored by adenosine triphosphate (ATP) molecules, which are both required and produced by metabolic pathways, and thus are referred to as cellular energy “currency”. Cells generate ATP through respiration, of which there are two distinct mechanisms: aerobic and anaerobic. Both of these pathways require the initial uptake of glucose, which is converted through a series of steps known as glycolysis to

pyruvate. However, at this point, what happens to pyruvate is typically dependent upon the environmental conditions surrounding the cell. Aerobic respiration, most often utilized by normal cells under normal, nonproliferating, conditions, requires oxygen and results in the conversion of the glycolytic product pyruvate to Acetyl Coenzyme A (acetyl-CoA). The primary function of Acetyl-CoA is to donate an acetyl group to the citric acid (also known as TCA or Krebs) cycle. By continuing on through the TCA cycle and electron transport chain (ETC) reactions, all of which take place in the mitochondria, the downstream metabolism of a single molecule of glucose by aerobic respiration yields a net gain of about 36 molecules of ATP, and releases carbon dioxide as a byproduct. This process is often collectively referred to as “oxidative phosphorylation” (Fig. 1A). Anaerobic respiration, on the other hand, is far less efficient, producing a net gain of only 2 molecules of ATP per molecule of glucose metabolized, and thus is typically only utilized during hypoxic or stressful conditions, as it does not require the presence of oxygen. During anaerobic respiration, also referred to as fermentation, pyruvate is converted to lactate and ethyl alcohol entirely within the cytosol. Although inefficient, this pathway can keep the cell alive during stressful conditions in which the supply of oxygen is low by generating enough ATP to continue sustained cycling through glycolysis (Fig. 1A).

While normal or quiescent cells rely primarily on aerobic respiration/ oxidative phosphorylation to meet their energy requirements, cancer cells appear to meet their increased demands for energy quite differently. Because tumor cells grow rapidly, they must increase the import of nutrients from their environment in an effort to maintain the pools of ATP, and even more importantly, carbon intermediates that serve as building blocks for the assembly of DNA, proteins, and lipids needed during cell growth and division. In the 1920's, Otto Warburg first made the observation that tumors took up markedly higher levels of glucose in comparison to normal tissues¹. Furthermore, he showed that even in the presence of ample oxygen, cancer cells produced much more lactate than normal tissues, suggesting that these cells were shuttling glucose through the glycolytic fermentation pathway². The sustained use of this pathway to meet energy requirements under normoxic conditions is now termed “aerobic glycolysis”, and the increased dependence on this pathway by cancer cells has come to be known as the “Warburg Effect” (Fig. 1A). In more recent years, this phenomenon has been confirmed in a number of different tumor types in different tissues, and has proven useful for diagnostic imaging using ¹⁸F-deoxyglucose positron emission tomography (FDG-PET) to detect the higher levels of glucose uptake observed in tumors in comparison to surrounding normal stroma³. It seems counter-intuitive that rapidly dividing cancer cells would prefer the less-efficient glycolytic pathway for meeting their energy demands. For this reason, Otto Warburg originally hypothesized that the increased rates of aerobic glycolysis in cancer cells were due to impaired function of the mitochondria in these cells, requiring them to rely solely upon glycolysis to make ATP needed for survival^{2,4}. This theory has been disproven in more recent years, however, as the majority of cancer cells have been found to maintain functioning mitochondria⁵. It has also become increasingly clear that tumor cells continue to carry out oxidative respiration in addition to sustained aerobic glycolysis (Fig. 1B), and that a likely advantage of this altered metabolic profile is the sustained production of glycolytic carbon intermediates required for the production of macromolecules needed by the rapidly dividing cells⁶.

While the Warburg Effect is perhaps the most recognized metabolic characteristic of many cancer cells, a broad range of metabolic alterations have been observed in tumors. In addition to increased glucose uptake, tumor cells have also been commonly shown to have higher levels of dependence on glutamine, which is a source of nitrogen for the synthesis of nucleotides and amino acids⁷. Interactions involving various intermediates of glycolysis, the TCA cycle, the ETC, and the Pentose Phosphate Pathway (PPP), as well as lipid metabolism pathways have all been shown to be altered in tumor cells and to play a role in tumorigenesis⁸. These metabolic changes can result from genetic aberrations in metabolic enzymes themselves, but can also be a downstream consequence of activating mutations in numerous growth factors and oncogenes, loss of tumor suppressor signaling, or epigenetic alterations⁷, all of which we will discuss in more detail in later sections of this manuscript. Recent findings demonstrating the influence of metabolic pathways on tumor cell proliferation, growth, and differentiation has renewed interest in identifying susceptibilities of these pathways to therapeutic intervention, and thus the investigation of metabolic reprogramming as a hallmark of cancer has become an extremely active area of research in the last decade³.

Metabolic Alterations In Renal Cell Carcinomas

The Renal Cell Carcinomas (RCCs) are prime examples of tumor types that are highly linked to alterations in metabolic pathways. There are three main subtypes of renal cell carcinoma: clear cell (ccRCC), papillary (pRCC), and chromophone (chRCC), each distinguished by unique histology and driver mutations⁹. Interestingly, although the overall mutational burden is relatively low in RCC in comparison to many other tumor types⁹, the vast majority of mutations identified in these tumors are in some way involved in the cell's ability to sense or respond to nutrients, oxygen, iron, or energy, suggesting that metabolic pathway alterations are key drivers of proliferation in all subsets of RCC¹⁰. Mutations resulting in dysregulation of specific steps of glycolysis, the TCA cycle, and the ETC pathways have all been found in subtypes of RCC, illustrating the diversity of metabolic alterations that may contribute to tumorigenesis (Fig. 2). Here, we discuss three examples of mutations that alter different metabolic pathways in RCC.

VHL Mutations in Clear Cell RCC

Mutations in the von-Hippel-Lindau gene (*VHL*) are associated with a hereditary form of RCC found in patients with germline VHL Disease, but are also observed in nearly 90% of patients with sporadic clear cell kidney cancer (ccRCC)¹¹. The VHL protein is considered a tumor suppressor, and under normal circumstances, when there is enough oxygen and iron in the cell, it is part of a complex that binds to the hypoxia-inducible factors (HIFs), and targets them for degradation by ubiquitination¹². In the majority of cases of ccRCC, inactivating mutations in *VHL* inhibit its ability to interact with the HIF proteins, and consequently the HIF proteins are stabilized, even during normoxic conditions. The HIF proteins are transcription factors that regulate the activity of a number of downstream genes, including glucose transporters *GLUT1* and *GLUT3*, endothelial growth factor (*EGF*), vascular endothelial growth factor (*VEGF*), and platelet-derived growth factor (*PDGF*)¹⁰. The aberrant activation of these proteins and growth factors is believed to contribute to tumor

growth and proliferation downstream of inactivating mutations in *VHL*. The upregulation of *GLUT1* and *GLUT3* likely contributes to the faster rates and increased levels of glycolysis in these tumors. A number of glycolytic enzymes are also transcriptionally regulated by HIFs, including *HK1*, *HK2*, *GPI*, *PFKL*, *ALDA*, *ALDC*, *TPI*, *GAPDH*, *PGK*, *ENO1*, and *PKM*. Increased expression of these enzymes may also contribute to the increased glycolytic activity in the *VHL*-mutant ccRCC tumors¹³. Finally, lactate dehydrogenase A (*LDHA*) expression is also transcriptionally regulated by HIFs. This enzyme converts the glycolytic product pyruvate to lactate, and thus upregulation of this enzyme contributes to the increased levels of lactic acid observed in tumors, a result of sustained aerobic glycolysis at the expense of the conversion of pyruvate to Acetyl-CoA for use in mitochondrial oxidative phosphorylation¹³. Thus, *VHL*-mutant tumors exhibit classic features of pseudo-hypoxic “Warburg” metabolism-upregulated glycolysis, high levels of lactate production, and lower levels of oxidative phosphorylation.

An understanding of how the VHL and HIF pathways contribute to ccRCC tumorigenesis has provided the basis for most current treatments of patients with advanced ccRCC. Most current therapies target the *VEGF* signaling pathway, inhibiting angiogenesis^{10,14}. Increased knowledge of the metabolic dependencies of RCC cells has also led to increased interest in targeting the HIF pathways and their metabolism-regulating targets. Recently, a HIF-2 agonist showed promise in reducing growth in a subset of ccRCC patient cell lines¹⁵. Agonists of GLUT1 and glycolytic pathway enzymes have also been investigated as potential therapeutic inhibitors of glycolysis in RCC^{16,17}. Further characterization of the metabolic reprogramming that occurs in ccRCC has the potential to identify additional vulnerabilities of therapeutic value.

***SDH* and *FH* Mutations in Papillary RCC**

Mutations in several TCA cycle enzymes have been observed in papillary renal cell carcinoma (pRCC). Succinate dehydrogenase (SDH) catalyzes the oxidation of succinate to fumarate in the TCA cycle. Germline mutations in the succinate dehydrogenase family subunits *SDHB*, *SDHC*, and *SDHD*, have been identified in patients with familial paraganglioma/pheochromocytoma, who are predisposed to developing pRCC tumors, and in other patients with a family history of pRCC¹⁸. Likewise, germline mutations in fumarate hydratase (FH), the enzyme that catalyzes the conversion of fumarate to malate in the TCA cycle, have been found in patients with Hereditary Leiomyomatosis Renal Cell Carcinoma (HLRCC) and very rarely, in sporadic cases of pRCC¹⁹. Since both *SDH* and *FH* mutations block normal TCA Cycle and ETC activity, cells from these tumors take up almost no oxygen, and rely primarily on glycolysis to supply energy and macromolecules needed for replication and growth. These tumors thus also exhibit “Warburg” metabolism, and produce high levels of lactate¹⁰.

In the case of *SDH*-deficient tumors, succinate accumulates in the mitochondrial matrix due to loss of SDH function. Succinate, however, can also leak out into the cytosol, where it can inhibit the prolyl hydroxylation of HIF complexes, preventing them from being targeted for proteasomal degradation. In this way, succinate accumulation stabilizes the HIF transcription factors, thus promoting the activation of their downstream targets, creating a pseudo-hypoxic

expression signature²⁰. Therefore, in addition to defective mitochondrial respiration, *SDH*-mutant cells also have increased expression of GLUT1, VEGF, and other growth factors and glycolytic enzymes, promoting cell growth and proliferation, angiogenesis, and means for upregulating glycolysis. These tumors have also been shown to have increased vasculature¹⁸. Although it has not yet been well-investigated due to the rarity of this tumor type, targeting the HIFs or the glycolytic pathway in these cells may have potential therapeutic value in these tumors¹⁰.

FH mutations similarly result in the accumulation of both succinate and fumarate due to the malfunction of the FH enzyme in the TCA cycle. Like succinate, fumarate can also move from the mitochondria into the cytoplasm, where it can interact with prolyl hydroxylases and prevent the degradation of HIF proteins²¹. Similarly to *SDH*-mutant tumors, pRCC tumors with *FH* mutations have upregulated expression of the HIF target genes involved in proliferation, glycolysis, and angiogenesis. Highly vascularized, these tumors grow very aggressively and have a pseudo-hypoxic gene expression profile²². Patients with these tumors typically have a poor prognosis, and more research is needed to identify improved therapies. The malfunctions of mitochondrial respiration and upregulation of glycolysis in these cells appear to be key factors in their proliferation, and thus investigation of these pathways may be important for improving outcome for patients with *FH* mutations.

ETC-I mutations in Chromophobe RCC

A third subset of RCC, known as chromophobe (chRCC), is the least common type of RCC. Like many of the renal cell carcinomas, this type of tumor is associated with a hereditary disorder, Birt-Hogg-Dube' (BHD) syndrome. Until more recent years, however, it was not known what genetic alterations contributed to sporadic cases of chRCC. Interestingly, PET/CT scans have demonstrated that, in contrast to other types of RCC, chRCC tumors are non-glycolytic, taking up very limited amounts of glucose²³. In addition, gene expression profiling of these tumors indicated that genes involved in the TCA Cycle and ETC pathways were upregulated in these tumors²⁴. Mitochondrial DNA sequencing has revealed that many chRCC tumors have mutations in genes involved in the ETC complex I, particularly in *MT-ND5*, and that these mitochondrial gene mutations also correlate with samples exhibiting an eosinophilic histological phenotype²⁴. This phenotype also correlates with an increase in mitochondrial mass resulting from an accumulation of mitochondria, possibly in compensation for hindered mitochondrial functioning⁹. Thus, the metabolic profile of chRCC appears to be very different from that of other types of kidney cancer. While the mechanisms behind the accumulation of mitochondria in this tumor type remains to be investigated, it is clear that metabolic alterations may play an important role in growth of this rare tumor type, and hence further study of these pathways for potential use as biomarkers and therapeutic targeting is warranted.

In summary, the renal cell carcinomas provide an illustration of the varied strategies employed by cancer cells to augment growth through manipulations of their metabolic activities. These activities reveal possible critical dependencies, which as has been referenced above, has been examined in terms of using altered glycolysis for diagnostic as well as potentially therapeutic intervention. Below, we will highlight the emerging strategies

to intervene in cellular metabolism for therapeutic benefit, including strategies currently approved in renal cancers and other malignancies, and new concepts that may apply in the future alone, or as adjuncts to treatments.

Other Metabolic Alterations In Cancer: Current Therapeutic Targets

A number of different kinds of genetic mutations have been associated with the dysregulation of metabolic pathways in various tumor types. The reprogramming of metabolism and alterations in metabolic flux of tumor cells compared to normal cells confers unique properties to these cells which may prove to be useful for therapeutic targeting in cancer patients. Here, we describe some of the pathways currently identified as regulators of metabolism in tumors and the current therapies targeting these alterations.

mTOR Inhibition

The mechanistic (previously mammalian) target of Rapamycin (mTOR) is a serine-threonine protein kinase that forms complexes with other proteins and is involved in a number of cellular processes related to growth, proliferation, survival, motility, and protein translation²⁵. mTOR signaling is commonly dysregulated in cancer through several different mechanisms. While mutations in the *MTOR* gene itself can occur, it is more commonly activated downstream of gain-of-function mutations in the PI3K-AKT pathway or growth factors, or through inactivation of tumor suppressors such as PTEN. mTOR is also activated downstream of activation of 5'-adenosine monophosphate-activated protein kinase (AMPK), a protein which serves as an intracellular sensor of nutrients²⁶. mTOR activation plays a key role in controlling intracellular metabolism through its involvement in protein translation and autophagy. The mTOR pathway has been shown to stimulate glutaminolysis by upregulating the expression of MYC which in turn upregulates glutaminase (GLS), which converts glutamine to glutamate that can be used in to make alpha ketoglutarate for use in the TCA cycle²⁷. mTOR activation is also known to play a role in the stabilization of HIF proteins, resulting in increased activation of their transcriptional targets, including GLUT1, VEGF, and other glycolysis enzymes²⁸. Thus, activation of the mTOR pathway plays a role in upregulation of glycolysis, glutamine uptake, and angiogenesis in cancer cells.

Two mTOR inhibitors, temsirolimus and everolimus, have been approved in the US and Europe for the treatment of solid tumors. These drugs bind to the mTOR complex 1 (mTORC1) by associating with FK506-binding protein12 (FKBP12), blocking the correct alignment of substrates to the catalytic cleft of this complex²⁹. These drugs have shown benefits in delaying the progression and extending survival in advanced renal cell carcinoma, breast, and pancreatic cancers. However, resistance to these inhibitors appears to develop over time, possibly due to the accumulation of additional mutations in the mTOR pathway³⁰ or through negative feedback of the pathway itself, as inhibiting mTOR signaling can also upregulate AKT signaling through insulin-like growth factor receptor 1 (IGF-1R)³¹. Thus, continued research to find less resistant mechanisms for inhibiting mTOR are needed.

Metformin/Phenformin Inhibition of Oxidative Phosphorylation

As cancer cells are frequently known to be metabolically active and have very high levels of glucose uptake, it has been postulated that hypo-glycemic drugs that have been used for treating diabetes could help to restore normal metabolism in these cells and prevent tumor growth. Two such drugs, Metformin and Phenformin, have shown some promise in targeting cancer cell metabolism. These organic compounds are known as biguanides, and it has been shown that diabetic patients taking them have a reduced risk of developing cancer^{32,33}. The exact mechanisms by which biguanides regulate cellular metabolism is not yet well understood, but they are believed to interfere with mitochondrial complex I, inhibiting oxidative phosphorylation, while activating the AMPK signaling pathway³⁴. Metformin and Phenformin have been shown to delay progression of tumor cell growth in breast cancer³⁵ and melanoma³², and have also exhibited anti-angiogenic properties³⁵. One disadvantage of treatment with these compounds is that they can induce severe acidosis in patients—more research is needed to determine the most effective dosage levels and which tumor types may be most susceptible to biguanide treatment.

Glutaminase Inhibition

While cancer cells have been shown to have highly upregulated glycolysis, demonstrated by the Warburg Effect, they also maintain oxidative phosphorylation. In addition to glycolysis, many cancer cells appear to be dependent upon glutamine metabolism to supply the nutrients and biosynthetic precursors that they need for macromolecule synthesis³⁶. The glutaminase enzyme (GLS) converts glutamine to glutamate, which can be used to make alpha-ketoglutarate, important in the TCA cycle. The TCA cycle intermediates are used in the synthesis of nucleic and fatty acids, and thus interfering with glutamine metabolism can have a profoundly detrimental effect on replicating cells.

Several mechanisms have been proposed for inhibiting glutaminase, including targeting ASCT2, the transporter that mediates glutamine uptake into cells, and the use of glutamine mimetics to competitively inhibit glutamine uptake and activity. Unfortunately, early clinical trials testing glutamine mimetics resulted in high levels of toxicity in patients³⁷. More recently, however, several small-molecule allosteric inhibitors of glutaminase activity have been identified, including CB-839, currently in clinical trials, and BPTES. Targeting glutaminase activity has been shown to reduce oncogenic transformation in cancer cells³⁸ and allosteric inhibitors of glutaminase have been used in combination with other chemotherapeutics to reduce tumor cell growth in lymphoma³⁹, lung⁴⁰, and breast⁴¹ cancer cell lines. Further research will likely focus on determining which tumors are most glutamine-dependent, and thus most susceptible to glutaminase inhibition. It will also be important to investigate the most efficient methods for targeting glutaminase activity in cancer cells while minimizing toxicity to others.

IDH1/2 Inhibition

The isocitrate dehydrogenase enzymes (IDH1 and IDH2) are important metabolic enzymes that convert isocitrate to alpha-ketoglutarate (α -KG) by oxidative decarboxylation. α -KG is a key player in the TCA cycle, and thus these enzymes play an important role in oxidative phosphorylation. IDH1 and 2 also play a role in the generation of NADPH, a reducing factor

that helps to protect the cell against oxidative damage⁴². Therefore, mutations in IDH1 and 2 are believed to both alter cellular metabolism and potentially increase rates of DNA damage due to altered NADPH protection. Mutations in IDH1 and 2 have been observed in several types of tumors, including leukemias, lymphomas, and gliomas. The mutations identified in IDH1 and 2 in cancers appear to be gain-of-function point mutations that occur at specific arginine residues that presumably alter the structure of these proteins. These mutations lead to increased conversion of α -KG to D-2-hydroxyglutarate (D-2HG)⁴³. High levels of D-2HG have been associated with increases in histone and DNA methylation, contributing to tumor progression⁴². It has also been shown that mutant IDH1 heterodimerizes with wild-type IDH1, inhibiting the activity of the wild-type enzyme and reducing levels of α -KG, which may play a role in the degradation of HIF proteins. Thus, mutant IDH1 may also play a role in the stabilization of HIFs and increased activation of their transcription factors involved in tumorigenesis and angiogenesis⁴³.

Several targeted chemical inhibitors of the activity of specific IDH1 and IDH2 point mutants have been designed and have been shown to reduce D-2HG and growth in cells and mouse models⁴³. Clinical trials using these inhibitors are ongoing in early stages. Another possible mechanism for inhibiting IDH1/2 signaling is to deprive them of α -KG using glutaminase inhibitors as described above⁴⁴. The study of IDH inhibition is ongoing in an effort to identify patients that may benefit from these therapies and which compounds and dosages are most effective.

Targeting Lipid Metabolism

While altered glucose and glutamine metabolism have been the primary focus of work studying the changes in metabolism of cancer cells, another aspect of cellular metabolism that is unique in proliferating and cancer cells is the oxidation and synthesis of lipids. Lipids such as fatty acids serve as an additional energy source for cells, and are required for membrane synthesis during cellular growth and division. Lipids can also play roles in cellular signaling by functioning as second messengers and as hormones⁴⁵. Fatty acids, the primary building block of cellular membranes, can be obtained from environmental sources or the cells can synthesize these molecules de-novo. Most normal adult cells prefer to get fatty acids from exogenous sources, but observations in cancer cells indicate that the de-novo synthesis of fatty acids is highly upregulated in many types of tumors⁴⁶. The shift in fatty acid synthesis in tumors has been suggested as a potential target to limit cancer cell growth.

One potential target for limiting lipid synthesis in tumor cells is the fatty acid synthase enzyme complex, FASN, which has also been found to be upregulated in some breast tumors⁴⁷. Currently, several chemical inhibitors, as well as genetic ablation of FASN by RNAi, are being tested for effectiveness in reducing tumor cell growth and proliferation⁴⁷. These studies have been extended to other enzymes involved in fatty acid synthesis as well. Other potential targets for inhibiting fatty acid synthesis are the sterol regulatory element-binding proteins (SREBPs), which are upstream regulators of lipid synthesis⁴⁷. So far, inhibitors of these molecules are in pre-clinical trial stages, as the investigation of potential side effects is necessary before they are given to patients.

Interactions Between Metabolism and Epigenetics

The metabolic reprogramming that occurs in cancer has far-reaching effects. In addition to altering metabolic pathways in response to nutrient uptake, metabolic changes also influence the epigenetic regulation of gene expression. Epigenetics are heritable changes in DNA that are not the result of an alteration in sequence, and include histone modifications such as methylation, acetylation, and phosphorylation. These epigenetic changes can influence gene expression by enhancing or repressing the transcription of genes. Thus, epigenetic changes downstream of metabolic alterations can influence the expression levels of many genes in cancer cells, possibly giving them a survival and growth advantage. In addition, the reverse could also be true—epigenetic alterations can influence cellular metabolism by altering the transcription of genes involved in metabolic pathways⁴⁸. These processes are tightly linked, and we will discuss several possible mechanisms for these interactions here.

The methylation of DNA at CpG sites in promoters is a mechanism by which epigenetic modifications repress the expression of genes. In cancer, DNA methylation is often observed in the promoter sites of tumor suppressor genes. DNA methylation is mediated by DNA methyltransferases (DNMTs), and histone methylation is mediated by histone methyltransferases (HMTs), both of which use an activated methyl donor from *S*-adenosylmethionine (SAM), a product of one-carbon metabolism. Dysregulation of carbon metabolism pathways in cancer can alter the levels of SAM and methyl donors available, thus influencing the epigenetic modifications and expression of genes in these cells⁴⁹.

Another mechanism by which metabolic pathways can effect epigenetics is through the TCA cycle metabolites. Several histone demethylases require the TCA cycle protein α -KG as a cofactor for activation, and thus the levels of TCA cycle intermediates may influence demethylase protein activity by competitive inhibition. Likewise, D-2HG, the product made from α -KG by cancer cells with IDH1/2 mutations (discussed above), inhibits the activity of α -KG-dependent demethylases. SDH and FH mutations which result in accumulation of succinate and fumarate in cancer cells can also act as competitive antagonists for inhibiting these α -KG-dependent demethylases. Thus, inhibition of demethylases in cancer cells by TCA cycle intermediates can result in hypermethylation of a variety of genes⁴⁹, contributing to the repression of tumor suppressor genes and others.

Another metabolic molecule that contributes to epigenetic programming is Acetyl-CoA. Acetyl-CoA fuels the TCA cycle and is involved in nearly all aspects of cellular metabolism, but is also used as a cofactor by enzymes that transfer acetyl groups, including histone acetyltransferases (HATs), which catalyze the addition of an acetyl group to histones. Histone acetylation is associated with transcriptional activation of genes. Thus, the availability of Acetyl-CoA, highly influenced by cellular metabolism pathways, also plays a role in the epigenetic regulation of gene expression^{48,49}.

These are just a few examples of ways in which metabolic alterations can influence the epigenetic regulation of gene expression. Thus, it must be considered that targeting metabolic pathways can also alter the epigenetic control of gene expression. Likewise, targeting epigenetic modification pathways also holds potential to alter gene expression,

including that of metabolism pathway enzymes. Future research investigating the links between epigenetics and metabolism will hopefully provide greater understanding of the complexity of the interactions between metabolism and chromatin dynamics in both normal and cancer cells.

Conclusions

Proliferating cancer cells must maintain both sufficient energy and pools of metabolic intermediates for building macromolecules needed for proliferation, including DNA, proteins, and lipids. These tasks are accomplished in most cancer cells by adapting their metabolism to be more dependent upon aerobic glycolysis and glutaminolysis. The mechanisms behind the metabolic reprogramming that takes place in most tumor cells are diverse, and include oncogenic activation, the repression of tumor suppressor signaling, epigenetic modifications, and mutations in metabolic enzymes themselves. As the metabolic profiles of tumor cells distinguish them from normal cells and are critical for their growth and survival, the metabolic signaling pathways have become desirable targets for therapeutic intervention in cancer patients. Recent work has focused on identifying inhibitors of critical metabolism pathways, and show promise in targeting metabolism to improve patient outcomes, either alone or in combination with other targeted therapies.

References

1. Warburg OP, Negelein E. Über den Stoffwechsel der Carcinomzelle. *Biochem Zeitschr.* 1927; 152:309–344.
2. Warburg O. On the Origin of Cancer Cells. *Science* (80-). 1956; 123
3. Ward PS, Thompson CB. Metabolic Reprogramming: A Cancer Hallmark Even Warburg Did Not Anticipate. *Cancer Cell.* 2012; 21:297–308. [PubMed: 22439925]
4. Weinhouse S, Warburg O, Burk D, Schade AL. On Respiratory Impairment in Cancer Cells. *Science* (80-). 1956; 124
5. Martínez-Reyes I, et al. TCA Cycle and Mitochondrial Membrane Potential Are Necessary for Diverse Biological Functions. *Mol Cell.* 2016; 61:199–209. [PubMed: 26725009]
6. Dang CV. Links between metabolism and cancer. *Genes Dev.* 2012; 26:877–90. [PubMed: 22549953]
7. Pavlova NN, Thompson CB. The Emerging Hallmarks of Cancer Metabolism. 2016; doi: 10.1016/j.cmet.2015.12.006
8. Cantor JR, Sabatini DM, Liations A. Cancer Cell Metabolism: One Hallmark, Many Faces. 2012; doi: 10.1158/2159-8290.CD-12-0345
9. Haake SM, Weyandt JD, Rathmell WK. Insights into the Genetic Basis of the Renal Cell Carcinomas from The Cancer Genome Atlas. *Mol Cancer Res.* 2016; 14
10. Linehan WM, Ricketts CJ. The metabolic basis of kidney cancer. *Semin Cancer Biol.* 2013; 23:46–55. [PubMed: 22705279]
11. Moore LE, et al. Von Hippel-Lindau (VHL) Inactivation in Sporadic Clear Cell Renal Cancer: Associations with Germline VHL Polymorphisms and Etiologic Risk Factors. *PLoS Genet.* 2011; 7:e1002312. [PubMed: 22022277]
12. Gossage L, Eisen T, Maher ER. VHL, the story of a tumour suppressor gene. *Nat Rev Cancer.* 2014; 15:55–64.
13. Semenza GL. HIF-1 mediates the Warburg effect in clear cell renal carcinoma. 2007; doi: 10.1007/s10863I007I9081I2
14. Jonasch E, et al. State of the science: an update on renal cell carcinoma. *Mol Cancer Res.* 2012; 10:859–80. [PubMed: 22638109]

15. Chen W, et al. Targeting renal cell carcinoma with a HIF-2 antagonist. *Nature*. 2016; 539:112–117. [PubMed: 27595394]
16. Chan DA, et al. Targeting GLUT1 and the Warburg Effect in Renal Cell Carcinoma by Chemical Synthetic Lethality. *Sci Transl Med*. 2011; 3
17. Van Der Mijn JC, Panka DJ, Geissler AK, Verheul HM, Mier JW. Novel drugs that target the metabolic reprogramming in renal cell cancer.
18. Bardella C, Pollard PJ, Tomlinson I. SDH mutations in cancer. *Biochim Biophys Acta - Bioenerg*. 2011; 1807:1432–1443.
19. Trpkov K, et al. Fumarate Hydratase-deficient Renal Cell Carcinoma Is Strongly Correlated With Fumarate Hydratase Mutation and Hereditary Leiomyomatosis and Renal Cell Carcinoma Syndrome. *Am J Surg Pathol*. 2016; 40:865–875. [PubMed: 26900816]
20. Selak MA, et al. Succinate links TCA cycle dysfunction to oncogenesis by inhibiting HIF-prolyl hydroxylase.
21. Sudarshan S, et al. Fumarate Hydratase Deficiency in Renal Cancer Induces Glycolytic Addiction and Hypoxia-Inducible Transcription Factor 1 Stabilization by Glucose-Dependent Generation of Reactive Oxygen Species. *Mol Cell Biol*. 2009; 29:4080–4090. [PubMed: 19470762]
22. King A, Selak MA, Gottlieb E. Succinate dehydrogenase and fumarate hydratase: linking mitochondrial dysfunction and cancer. *Oncogene*. 2006; 25:4675–4682. [PubMed: 16892081]
23. Rathmell KW, Chen F, Creighton CJ. Genomics of chromophobe renal cell carcinoma: implications from a rare tumor for pan-cancer studies. *Oncoscience*. 2015; 2
24. Davis CF, et al. The Somatic Genomic Landscape of Chromophobe Renal Cell Carcinoma. *Cancer Cell*. 2014; 26:319–330. [PubMed: 25155756]
25. Laplante M, Sabatini DM. mTOR signaling at a glance. *J Cell Sci*. 122:3589–3594.
26. Kennedy BK, Lamming DW. The Mechanistic Target of Rapamycin: The Grand Conductor of Metabolism and Aging. 2016; doi: 10.1016/j.cmet.2016.05.009
27. Jin L, Alesi GN, Kang S. Glutaminolysis as a target for cancer therapy. *Oncogene*. 2016; 35:3619–3625. [PubMed: 26592449]
28. Land SC, Tee AR. Hypoxia-inducible factor 1alpha is regulated by the mammalian target of rapamycin (mTOR) via an mTOR signaling motif. *J Biol Chem*. 2007; 282:20534–43. [PubMed: 17502379]
29. Weichhart T, Hengstschläger M, Linke M. Regulation of innate immune cell function by mTOR. *Nat Rev Immunol*. 2015; 15:599–614. [PubMed: 26403194]
30. Grabiner BC, et al. A Diverse Array of Cancer-Associated MTOR Mutations Are Hyperactivating and Can Predict Rapamycin Sensitivity. *Cancer Discov*. 2014; 4
31. Wan X, Harkavy B, Shen N, Grohar P, Helman LJ. Rapamycin induces feedback activation of Akt signaling through an IGF-1R-dependent mechanism. *Oncogene*. 2007; 26:1932–1940. [PubMed: 17001314]
32. Petrachi T, et al. Therapeutic potential of the metabolic modulator phenformin in targeting the stem cell compartment in melanoma. *Oncotarget*. 2016; 8:6914–6928.
33. Liu Z, et al. Phenformin Induces Cell Cycle Change, Apoptosis, and Mesenchymal-Epithelial Transition and Regulates the AMPK/mTOR/p70s6k and MAPK/ERK Pathways in Breast Cancer Cells.
34. Hur KY, Lee MS. New mechanisms of metformin action: Focusing on mitochondria and the gut. *J Diabetes Investig*. 2015; 6:600–609.
35. Orecchioni S, et al. The biguanides metformin and phenformin inhibit angiogenesis, local and metastatic growth of breast cancer by targeting both neoplastic and microenvironment cells. *Int J Cancer*. 2015; 136:E534–E544. [PubMed: 25196138]
36. Deberardinis RJ, Chandel NS. Fundamentals of cancer metabolism INTRODUCTION AND OVERARCHING PRINCIPLES.
37. Lukey MJ, Wilson KF, Cerione RA. Therapeutic strategies impacting cancer cell glutamine metabolism. *Future Med Chem*. 2013; 5:1685–700. [PubMed: 24047273]
38. Wang JB, et al. Targeting Mitochondrial Glutaminase Activity Inhibits Oncogenic Transformation. *Cancer Cell*. 2010; 18:207–219. [PubMed: 20832749]

39. Xiang Y, et al. Targeted inhibition of tumor-specific glutaminase diminishes cell-autonomous tumorigenesis. *J Clin Invest*. 2015; 125:2293–2306. [PubMed: 25915584]
40. Momcilovic M, et al. Targeted Inhibition of EGFR and Glutaminase Induces Metabolic Crisis in EGFR Mutant Lung Cancer. *Cell Reports*. 2017; 18
41. Gross MI, et al. Antitumor Activity of the Glutaminase Inhibitor CB-839 in Triple-Negative Breast Cancer. *Mol Cancer Ther*. 2014; 13
42. Dang L, Yen K, Attar EC. IDH mutations in cancer and progress toward development of targeted therapeutics. *Ann Oncol*. 2016; 27:599–608. [PubMed: 27005468]
43. Fujii T, Khawaja MR, DiNardo CD, Atkins JT, Janku F. Targeting Isocitrate Dehydrogenase (IDH) in Cancer. *Discov Med*. 21:373–380.
44. Laurenti G, Tennant DA. Isocitrate dehydrogenase (IDH), succinate dehydrogenase (SDH), fumarate hydratase (FH): three players for one phenotype in cancer? *Mitochondria Heal Dis*.
45. Santos CR, Schulze A. Lipid metabolism in cancer. *FEBS J*. 2012; 279:2610–2623. [PubMed: 22621751]
46. Currie E, Schulze A, Zechner R, Walther TC, Farese RV. Cellular Fatty Acid Metabolism and Cancer. *Cell Metab*. 2013; 18:153–161. [PubMed: 23791484]
47. Röhrig F, Schulze A. The multifaceted roles of fatty acid synthesis in cancer. *Nat Rev Cancer*. 2016; 16:732–749. [PubMed: 27658529]
48. Lu C, Thompson CB. Metabolic Regulation of Epigenetics. 2012; doi: 10.1016/j.cmet.2012.06.001
49. Wong C, Qian Y, Yu J. Interplay between epigenetics and metabolism in oncogenesis: mechanisms and therapeutic approaches. *Nat Publ Gr*. 2016; doi: 10.1038/onc.2016.485

Figure 1. Metabolic Reprogramming In Tumor Cells: The Warburg Effect

(A.) Under normoxic conditions, normal tissues convert the glycolytic product of pyruvate to Acetyl-CoA, which is used in the mitochondria for the TCA Cycle to begin the process of oxidative phosphorylation. In the absence of oxygen, pyruvate is converted to lactate and sustained anaerobic glycolysis is used to meet requirements for energy and nutrients.

(B.) Tumor cells convert the majority of the glycolytic product pyruvate to lactate and replenish their nutrients and energy through sustained aerobic glycolysis., but maintain mitochondrial function and some oxidative respiration.

Figure 2. Metabolic Alterations In Renal Cell Carcinoma Subsets

Clear Cell RCC (blue background) frequently exhibits mutations in *VHL*, resulting in stabilization of HIFs and their transcriptional targets, including VEGF and GLUT1, and thus is characterized by increased angiogenesis and upregulated glycolysis. Mutations in *FH* and *SDH* in Papillary RCC (green background), inhibit completion of the TCA cycle and result in accumulation of fumarate and/or succinate. Chromophobe RCC (pink background) is rare, but associated with mutations in mitochondrial complex I enzymes, such as *MT-ND5*, leading to an inhibition of electron transport chain reactions and an accumulation of defective mitochondria.