Review Article

Unravelling the connection between metabolism and tumorigenesis through studies of the liver kinase B1 tumour suppressor

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Abstract

The liver kinase B1 (LKB1) tumour suppressor functions as a master regulator of growth, metabolism and survival in cells, which is frequently mutated in sporadic human non-small cell lung and cervical cancers. LKB1 functions as a key upstream activator of the AMP-activated protein kinase (AMPK), a central metabolic switch found in all eukaryotes that govern glucose and lipid metabolism and autophagy in response to alterations in nutrients and intracellular energy levels. The LKB1/AMPK signalling pathway suppresses mammalian target of rapamycin complex 1 (mTORC1), an essential regulator of cell growth in all eukaryotes that is deregulated in a majority of human cancers. LKB1 inactivation in cancer leads to both tumorigenesis and metabolic deregulation through the AMPK and mTORC1-signalling axis and there remain critical challenges to elucidate the direct role LKB1 inactivation plays in driving aberrant metabolism and tumour growth. This review addresses past and current efforts to delineate the molecular mechanisms fueling metabolic deregulation and tumorigenesis following LKB1 inactivation as well as translational promise of therapeutic strategies aimed at targeting LKB1-deficient tumors.

Keywords: AMP-activated protein kinase, fluoro-deoxyglucose positron emission tomography, liver kinase B1, mammalian target of rapamycin complex 1, therapeutics, Warburg

INTRODUCTION

The liver kinase B1 (LKB1) tumour suppressor (LKB1 also known as serine/threonine kinase 11) that functions as a master regulator of growth, metabolism and survival in cells. LKB1 was originally identified as a tumour suppressor gene located on chromosome 19p13 where loss or mutation of this gene is responsible for the inherited cancer disorder Peutz-Jeghers Syndrome (PJS).[1] Importantly, LKB1 is frequently mutated in ~30% of sporadic human non-small cell lung cancer (NSCLC) and 20% of cervical cancers.[2-4] A critical link directly connecting LKB1 to cell metabolism and cancer came from the discovery that LKB1 was the key upstream activator of the AMP-activated protein kinase (AMPK),[5-8] a central metabolic switch found in all eukaryotes that governs glucose and lipid metabolism and autophagy in response to alterations in nutrients and intracellular energy levels. In addition, LKB1 functions as a negative regulator of mammalian target of rapamycin complex 1 (mTORC1), an essential integrator of nutrient and growth factor signals that controls cell growth in all eukaryotes and is deregulated in a majority of human cancers.[9] Nearly 90 years ago Otto Warburg first

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10.4103/1477-3163.116323</td></tr>
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described aerobic glycolysis in tumour cells and renewed interest in the Warburg effect has propelled metabolism to the forefront of cancer biology.[10] LKB1 inactivation in cancer leads to both tumorigenesis and metabolic deregulation through the AMPK and mTORC1-signalling axis and there remain critical challenges to elucidate the direct role LKB1 inactivation plays in driving aberrant metabolism and tumour growth. This review will address past and current efforts to delineate the molecular mechanisms driving metabolic deregulation and tumorigenesis following LKB1 inactivation as well as therapeutic strategies to target LKB1 regulated pathways and their translational promise.

LKB1 IS A MASTER KINASE REGULATING METABOLISM AND GROWTH THROUGH AMPK AND MTOR SIGNALLING

LKB1 regulation of growth and metabolism through AMPK and mTOR signalling

The activated LKB1 kinase exists as a trimeric complex with the pseudo kinase STE20-related adaptor-α STRADα) and scaffolding protein mouse protein 25α,[11-13] where it regulates metabolism through the direct phosphorylation and activation of AMPK along with 12 other closely related AMPK-like kinases whose functions span a broad spectrum of biology including regulation of metabolism, growth, polarity and cell survival.[14,15] AMPK is the only LKB1 substrate that is activated under low adenosine triphosphate (ATP) conditions following nutrient deprivation or hypoxia and functions as a cellular rheostat maintaining energy homeostasis. AMPK exists as a heterotrimeric complex composed of a catalytic subunit (AMPKα1, α2) and two regulatory subunits (AMPKβ1, β2 and AMPKγ1, γ2, γ3). AMPK is activated upon the direct binding of adenosine diphosphate (ADP) or adenosine monophosphate (AMP) to the γ subunit where AMPK undergoes a conformational change leading to the phosphorylation of Thr172 on the activation loop of the α subunit.[16-18] LKB1 was found to be the key upstream activating kinase of AMPK,[6-8] however, AMPK is activated as weel by calmodulin-dependent protein kinase kinase β in response to calcium flux and potentially TAK1 in certain cellular contexts.[19-21]

During periods of energetic stress AMPK activation results in the activation of catabolic pathways and the concomitant inhibition of anabolic metabolism, which serve to restore energy homeostasis. Acute activation of AMPK regulates cellular metabolism through activation of AMPK substrates including the 6-phosphofructo-2-kinase pathway, acetyl coenzyme A carboxylase, 3-hydroxy-3-methyl coenzyme A reductase, which rapidly lead to the activation of glucose metabolism and fatty acid synthesis.[22-25] Long-term activation

of AMPK results in the phosphorylation and activation of transcriptional factors involved in adaptive reprogramming of cellular metabolism. For complete and detailed reviews of AMPK's role in regulation of metabolism refer to reviews by Mihaylova and Shaw 2011 and Hardie *et al.*, 2012.[26,27]

The LKB1/AMPK signalling pathway is a negative regulator of the mTORC1 signalling complex [Figure 1], a central integrator of nutrient and growth factor inputs that controls growth and the metabolic landscape of cells in all eukaryotes and is deregulated in a majority of human cancers,[28,29] mTOR is found in two biochemically and functionally discrete signalling complexes,[30] mTOR complex 1 includes raptor, which acts as a scaffold to recruit downstream substrates such as translation initiation factor 4E-binding protein 1 (4E-BP1) and ribosomal S6 kinase (p70S6K1) that contribute to mTORC1-dependent regulation of protein translation.[31] In contrast, mTOR complex 2 contains the rictor subunit and is neither sensitive to nutrients nor acutely inhibited by rapamycin,[9] mTORC1 controls the translation of a number of cell growth regulators, including cyclin D1, hypoxia inducible factor 1 α (HIF-1 α) and c-Myc, which in turn promote processes including cell cycle progression, cell growth, metabolism and angiogenesis, all of which can become deregulated during tumorigenesis.[9] AMPK inhibits mTORC1 through the direct phosphorylation of both tuberous sclerosis complex 2 (TSC2) and the mTORC1 scaffolding protein raptor.[32-35] TSC2 inhibits mTORC1 indirectly via regulation of the small guanine triphosphate hydrolase (GTPase) Rheb, such that loss of TSC1 or TSC2 leads to hyperactivation of mTORC1,[36] while phosphorylation of raptor was shown to be required for down regulation of mTORC1 and efficient G2/M cell cycle arrest following AMPK activation,[35] [Figure 1].

LKB1 INACTIVATION LEADS TO TUMORIGENESIS AND ALTERED METABOLISM

LKB1 is a frequently mutated tumour suppressor gene

Mutations in LKB1 lead to an inherited cancer syndrome known as PJS collectively belong to a group of syndromes known as phakmatoses in which patients develop benign hamartomas and are predisposed to a number of other malignancies, including breast, ovarian, endometrial and pancreatic tumors,[37] many of which have been studied in specific LKB1 mouse models [Table 1]. This increased cancer risk in PJS patients prompted the discovery that LKB1 is frequently mutated in lung and cervical cancer patients, which has intensified the focus to understanding the molecular basis of LKB1 mediated carcinogenesis.[2,4] LKB1 mutations are most
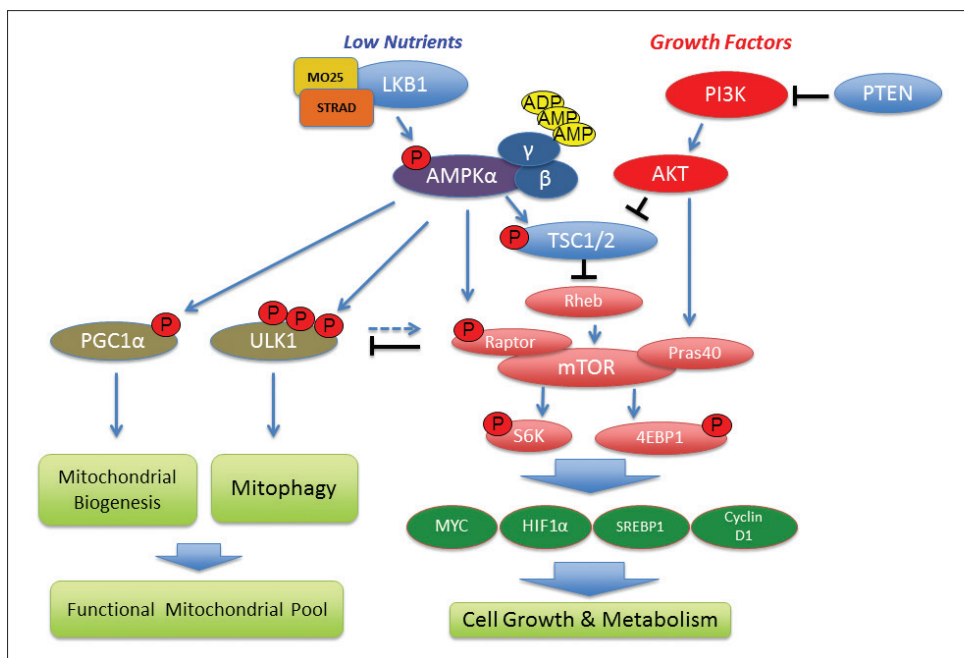
Figure 1: Schematic representation of the Liver Kinase B1 dependent regulation of growth, metabolism and mitochondrial homeostasis through AMP-activated protein kinase (AMPK) and mammalian target of rapamycin complex 1 (mTORC1) signalling pathways. Both tumour suppressors (in blue) and oncogenes (in red) regulate mTORC1 and are frequently mutated in cancer. mTORC1 effectors S6K and 4E-binding protein 1 and mTORC1-regulated transcription factors such as sterol regulatory element-binding protein 1 (SREBP1) and HIF1α, cMYC and Cyclin D1 (in green) are involved in cell growth, lipid and glucose metabolism and angiogenesis. AMPK suppresses mTORC1 through activation of tuberous sclerosis complex 2 and raptor and regulates mitochondrial homeostasis through peroxisome proliferator-activated receptor gamma coactivator 1-α (PGC1 α) and Unc51 like kinase 1

prevalent in male smokers and frequently co-mutate with the Kirsten rat sarcoma viral oncogene homolog (KRAS) and p53 genes, which may in part explain the low frequency of lung cancer in PJS patients as cigarette smoke likely compounds the mutational spectrum in NSCLC, however, definitive studies directly linking cigarette smoke to LKB1 mutation have yet to be performed. Importantly, LKB1 inactivation may be an early event in carcinogenesis as LKB1 mutations have been found in early as well as late stage primary lung tumors.[38] A recent study examining organ specific deletion of LKB1 during the mouse development found that LKB1 inactivation alone lead to growth of pancreatic lesions *ex vivo*.[39] Studies of LKB1 inactivation in genetically engineered mouse models (GEMMs) of disease further support the idea that loss of LKB1 may in fact be an early event in carcinogenesis sufficient to drive tumour growth. GEMMs targeting LKB1 inactivation in a broad spectrum of tissue accurately mimic human tumors commonly found to bear LKB1 mutations [Table 1].

LKB1-AMPK regulation of the Warburg effect *in vivo*
Deletion of LKB1 in human cancer cell lines and murine embryonic fibroblasts (MEFs) and gastrointestinal hamartomas from LKB1 +/- mice resulted in hyperactivation mTORC1.[34,40] Loss of function studies of LKB1 or AMPK in fibroblasts as well as epithelium from gastrointestinal hamartomas from Peutz-Jeghers patients or LKB1 +/- mice also show

increased expression results in increased HIF-1 α and its targets Glucose transporter 1 (GLUT1) and hexokinase in a rapamycin-reversible manner,[41] suggesting that HIF-1 α may be a relevant target downstream of LKB1-deficiency in PJS. 18F-Fluoro-deoxyglucose positron emission tomography ([18]FDG-PET) imaging studies on LKB1 +/- mice showed that their gastrointestinal hamartomas have increased maximum standard uptake values (SUVmax) correlating with the increased GLUT1 expression and suggest that loss of LKB1 drives glycolytic metabolism.[41]

Recent studies in murine T and B-cells and hematopoietic stems cells (HSCs) have demonstrated the LKB1-AMPK pathway is critical to metabolism and survival.[42-45] A recent loss of function study of AMPKα1 in the Eμ-Myc murine lymphoma model and in fibroblasts confirmed the role of the LKB1-AMPK pathway in suppressing the Warburg effect as deletion of AMPKα1 resulted in increased mTORC1 and HIF1 α dependent glucose metabolism. This is the first study to identify AMPK as a potential tumour suppressor gene as inactivation of AMPKα1 -/- resulted in accelerated tumour growth of Eμ-Myc lymphomas.[46] While both T and B cells only have one copy of AMPKα1,[47] the majority of human and murine epithelial tissues expresses both α1 and α2 genes and it will be important to identify the tumour suppressor functions of both the α1 and α2 genes in various tissues. Studies of LKB1

Table 1: LKB1 mutations in human disease and modeling Lkb1 inactivation in cancer using genetically engineered mouse models (GEMMs)

Human Disease	GEMMs	Phenotype	Refs
Peutz-Jeghers Syndrome	Lkb1+/- Lkb1+/-;p53+/- and Lkb1+/-;p53+/-SM22-Cre-Lkb1 $^{lox/lox}$	Benign intestinal hamartomas, accelerated intestinal hamartomas with p53 loss, osteoblastic differentiation	[40, 90-93]
Hepatocellular Carcinoma	Lkb1+/-; p53+/- Lkb1+/-; p53-/-	Hepatocellular carcinomas (in one mouse strain), with evidence of osteosarcomas and lymphomas	[94-96]
Non-Small Cell Lung Cancer	Lox-Stop-Lox-Kras ^{G12D}; Lkb1 $^{lox/lox}$ (delivered by intranasal inhalation of adeno-Cre virus)	Development of aggressive non-small cell lung carcinomas that have adeno, squamous large cell and mucinous adenocarcinoma in situ tumors; widespread metastases.	[2,3,97,98]
Cervical Cancer	Lkb1+/- or Lkb1 $^{lox/lox}$ (intrauterine injection of adeno-Cre virus) Sprr2f-Cre-Lkb1 $^{lox/lox}$ Misr2-Cre-Lkb1 $^{lox/lox}$ Lkb1 $^{lox/lox}$;Pten $^{lox/lox}$	Invasive endometrial adenocarcinoma; oviductal neoplasia	[4,56,57, 99]
Melanoma	Tyr-Cre-ER ^{T2}-Lox-Stop-Lox-Kras ^{G12D}; Lkb1 $^{lox/lox}$	Development of Aggressive and metastatic melanomas	[86]
Breast Cancer	BLG-Cre-Lkb1 $^{lox/lox}$ WAP-Cre-cMyc;Lkb1 $^{lox/lox}$	Mammary tumors, occasional DCIS and squamous differentiation. Accelerated development of mammary intraepithelial neoplasia (MIN) and adenocarcinoma	[100,101]
Prostate Cancer	P450CYP1A1-Cre-Lkb1 $^{lox/lox}$	Hyperplasia and neoplasia of the prostate epithelium	[102]
Pancreatic Cancer	Lkb1+/- or Pdx-Cre-Lkb1 $^{lox/lox}$ and Pdx-Cre-LSL-Kras ^{G12D}; Lkb1 $^{lox/+}$	Benign pancreatic cystadenomas Pancreatic ductal adenocarcinoma (PDAC)	[103] [106]
Head and neck squamous cell carcinoma	K14-Cre-Lkb1 $^{lox/lox}$ or Lkb1+/- with DMBA administered to skin	Squamous cell carcinoma of the skin and lung	[104,105]
Bladder	AhCreER ^{TM}-Lkb1 $^{lox/lox}$ AhCreER ^{TM}-Lkb1 $^{lox/lox}$;Pten $^{lox/lox}$	Papillary tumors Accelerated papillary tumors with squamous differentiation	[55]
Lymphoma	Lkb1 hypomorph;Pten $^{+/-}$ Lck-Cre-Lkb1 $^{lox/lox}$	Accelerated development of lymphomas Impaired T cell development	[42,80]

inactivation in NSCLC likewise revealed increased mTORC1 signalling and metabolic deregulation that correlated with elevated GLUT1 expression and increased SUVmax following ^{18}FDG-PET imaging in both human and murine lung tumors suggesting that increased glucose metabolism is a conserved phenotype in LKB1 -/- tumors.[48,49]

LKB1 mediated regulation of mitochondrial homeostasis

Recent work in HSCs and NSCLC has shed light on LKB1 role as a regulator of mitochondrial function and aerobic metabolism, where LKB1 -/- murine HSCs show mitochondrial defects including increased mitochondrial content and reduced mitochondrial membrane function.[43-45] It was recently discovered that AMPK directly phosphorylates the Unc51 like kinase (ULK1), a key upstream regulator of autophagy and mitochondrial turnover known as mitophagy.[50] AMPK -/- and ULK1 -/- MEFs displayed mitochondrial defects similar to LKB1 -/- HSCs and a loss of mitophagy resulting in the accumulation of defective mitochondria. Analysis of LKB1 -/- NSCLC tumour lines revealed mitochondrial defects and inactivation of the AMPK-ULK1 mitophagy signalling pathway as well as reduced oxidative phosphorylation.[48] These studies suggest that LKB1 inactivation and the resultant loss of AMPK-ULK1 regulated mitophagy may lead to aberrant mitochondrial pools compromising aerobic respiration in

tumour cells. As both mitophagy and biogenesis are regulated by AMPK through peroxisome proliferator-activated receptor gamma coactivator 1-α (PGC-1 α) and ULK1, the accumulation of defective mitochondria following LKB1 loss appears to reduce the respiratory capacity in tumour cells and may be another mechanism by which LKB1 inactivation drives glycolytic metabolism. Additionally, Myc overexpressing tumour cells were shown to rely on the LKB1 substrate AMPK-related protein kinase 5 (ARK5, also known as NUAK1) and the AMPK-mTORC1 signalling axis to maintain glutamine metabolism during energy stress and readily underwent apoptosis following glucose starvation or inhibition of ARK5 and mTORC1.[51] Future metabolomics studies investigating the integration of respiration, glycolysis and glutaminolysis will shed light to the global metabolic rewiring that occurs following LKB1 loss.

THERAPEUTICALLY TARGETING THE LKB1-AMPK SIGNALLING PATHWAY IN CANCER

Targeting mTOR signalling in LKB1-deficient tumors

Preclinical studies examining rapamycin in Lkb1 deficient murine models of PJS, lung and endometrial cancers have yielded mixed results. Spontaneously arising hamartomas

in Lkb1-/- mice responded well.[41,52] Translation of these rapamycin studies to treat phakmatoses in clinical trials has proven successful in TSC and Lymphangioleiomyomatosis patients,[53,54] while early phase clinical trial testing rapalogs in PJS patients have suffered from low enrollment. Rapamycin as a single agent has been shown to potently inhibit growth and viability of endometrial carcinomas, ovaductal neoplasias and papillary bladder tumors in Lkb1-/- and Lkb1-/-; Pten-/- GEMMs,[55-57] however, in Kras[G12D] driven LKB1 deficient GEMMs of lung cancer rapamycin failed to induce a therapeutic response.[58] Clinical trials assessing rapalogs on advanced stage tumors have seen variable results and much of the resistance of advanced tumors to rapamycin may be due to mTORC2-AKT mediated reactivation of mTORC1 and rapalogs have performed poorly in clinical trials as tumour frequently become resistant.[59] It will be important to identify the appropriate genetic and molecular context in advanced tumors that dictate either sensitivity or resistance to rapamycin and rapalogs.

The development of next generation mTOR inhibitors that target the kinase domain of mTOR or dual mTOR/PI3K may in fact induce a greater therapeutic response with targeted cytotoxicity to mTOR dependent tumors.[60-62] Treatment of KRAS driven, PI3KCA mutant murine lung tumors with the dual PI3K/mTOR inhibitor BEZ235 resulted in inhibition of both PI3K and mTOR signalling and regression of primary lung tumors. Importantly, inhibition of the PI3K/mTOR pathway, lead to a dramatic reduction of [18]FDG uptake suggesting a robust inhibition of glucose metabolism.[63] A recent study that examined the mTOR kinase inhibitor INK128 found this drug to dramatically inhibit mTORC1 and mTORC2 signalling in both human and murine PTEN -/- prostate tumour models that resulted in tumour regression and inhibition of epithelial-mesenchymal transition through down regulation of the mTOR-mediated translatome.[64] The predicted effectiveness of targeted mTOR kinase inhibitors against LKB1 deficient tumors of different tissues remains to be tested.

Using AMPK agonists as cancer therapeutics

AMPK's negative regulation of mTORC1 [Figure 1] and its ability to both acutely and durably rewire cellular metabolism suggest that AMPK activating drugs may be useful as cancer therapeutics.[65] One of the most commonly used AMPK agonist is the biguanide metformin (Glucophage), which is the most widely used type 2 diabetes drug in the world and taken by approximately 120 million patients daily. Several retrospective studies revealed a strong correlation between reduced cancer risk and mortality in diabetic patients taking metformin,[66-68] agreeing with early studies showing that biguanides suppressed naturally arising tumors in both transgenic and carcinogen treated rodent cancer models.[69,70]

Metformin and its more potent analog phenformin induce energy stress through inhibition of complex I of the mitochondrial respiratory chain, resulting in elevated intracellular ADP/AMP levels and AMPK activation.[71] Biguanides are thought exert their anti-cancer effects on tumour cells in part through activation of the AMPK pathway, but in reality these drugs work through a broad spectrum of mechanisms that extend beyond the AMPK pathway solely.[72]

AMPK agonists such as metformin, phenformin, AICAR, 2-deoxyglucose and thiazolidinediones have been shown to inhibit the growth of a wide variety of tumour cells in culture and xenografts in AMPK-dependent and independent manners,[73] for a detailed review please see reference.[74] Other widely used metabolic stress agents and AMPK activators include the natural product berberine found in green tea and salicylic acid (aspirin) found in most bathroom medicine cabinets. Recently, aspirin was identified as a direct AMPK activator,[75] and strikingly, colorectal cancer patients with PI3KCA mutations who took aspirin showed a significant reduction in mortality.[76] A separate study found increased AMPK activation and reduced mTORC1 signalling in colorectal cancer biopsies samples from patients taking aspirin daily,[77] suggesting that inhibition of PI3K/mTOR signalling via aspirin mediated activation of AMPK may lie at the heart of such striking results.

Inhibiting tumour cell growth and proliferation through AMPK activation in LKB1-/- tumour cells can also be achieved by AMPK agonists that work independently of LKB1. The small molecule Abbott A769662 is an allosteric activator of AMPK that can directly activate AMPK independently of LKB1, however, the exact region of the AMPK complex to which A769662 binds is unknown.[78,79] A769662 showed *in vivo* activation of AMPK and anti-tumour efficacy in a Pten +/- model of lymphoma.[80] Selective killing of LKB1 -/- tumour cells can also be achieved by exploiting metabolic and oxidative stress pathways that become deregulated upon inactivation of the LKB1/AMPK pathway. Tumour cells lacking LKB1 are hypersensitive to apoptosis in culture following treatment with energy stress inducing agents, presumably originating from an inability to restore ATP levels due to AMPK deficiency.[8] We have recently shown that LKB1 loss selectively sensitizes human and murine models of NSCLC to phenformin through metabolic and oxidative stress,[48] [Figure 2]. Our results agree with recent studies that showed AMPK mediates cell survival not only through maintaining cellular ATP but also by restoring nicotinamide adenine dinucleotide phosphate (NADPH) levels required to quench reactive oxygen species that accumulate during periods of metabolic stress.[83] Expanding the use of phenformin beyond LKB1 deficient lung tumors,
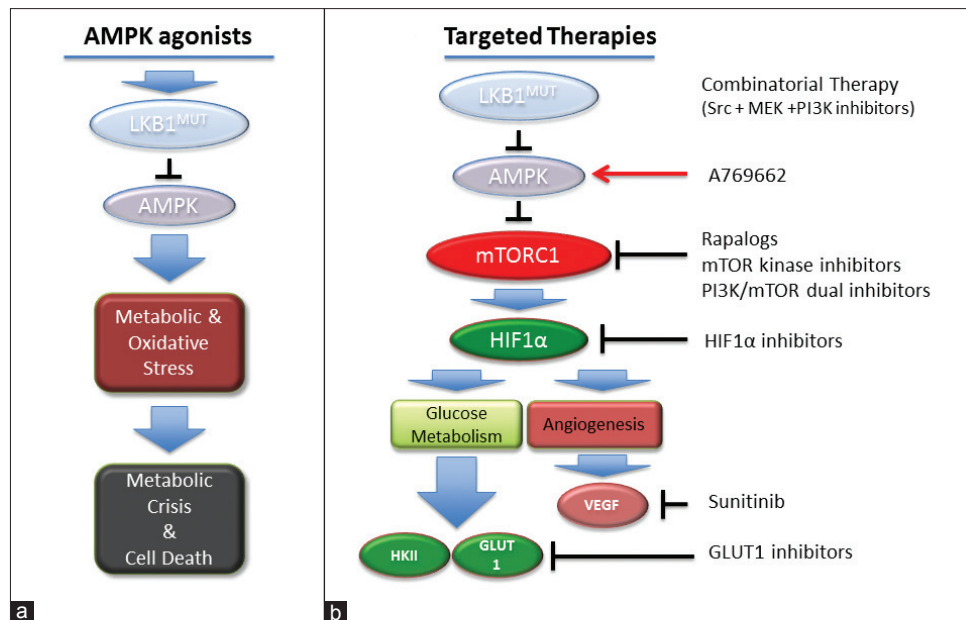
Figure 2: Therapeutically targeting the AMP-activated protein kinase (AMPK) and mammalian target of rapamycin complex (mTORC1) signaling pathways in liver kinase B1 (LKB1) -/- tumors. Schematic representation of therapeutic strategies targeting AMPK and the mTORC1 signaling pathway following LKB1 inactivation. (a) A representation of AMPK agonists that mimic cellular energy stress creating metabolic and oxidative stress that results in tumor cell death. (b) A representation of therapies targeting critical upstream and downstream effectors of mTORC1 in LKB1 -/- tumors

a recent study found that phenformin significantly inhibited growth of estrogen receptor positive and triple negative breast cancer cells in xenograft models.[81] Phenformin was withdrawn from clinical use in the late 1970s due to the occurance of lactic acidosis in a subset of diabetics on the drug,[82] however, it may find modern use as an anti-cancer agent as the dosing and duration of its use for cancer would be quite different from its use for diabetes.[84] Given the known pharmacokinetics and widespread long-term clinical use of biguanides, the potential for metabolic therapies to be repurposed as chemotherapies to target LKB1 deficient tumors warrants further attention.

Translational outlook – the promise and the reality

Before LKB1 -/- tumors can ever be successfully treated with targeted or metabolic therapies it must become common practice to perform genetic screening for LKB1 mutations in tumour types with a high frequency of LKB1 mutations. The elevated glycolytic metabolism of LKB1-deficient tumors in patients should allow these tumors to be readily imaged by [18]FDG-PET. [18]FDG uptake need not only be viewed as a surrogate end stage biomarker but as a diagnostic tool providing details to tumour metabolism and therapeutic response following treatment.[48,49] AMPK activating drugs and metabolic therapies, like most targeted therapeutics, are expected to be most effective against tumors of specific genotypes. The relative success of biguanides in preclinical studies must be taken cautiously as many studies are performed at supra-physiological concentrations in cell culture and

rodent cancer models that will never be achieved in man.[84] Therefore, careful studies of biguanides and other AMPK agonists at physiologically relevant concentrations are needed to understand their true potential. Given the multitude of deregulated effectors following LKB1 inactivation, it is likely that a combination of therapies will be required. Combining PI3K and MEK inhibitors with the Src inhibitor dasatinib in murine Kras[G12D]; Lkb1 -/- (KL) GEMMs induced a potent tumour response in non-small cell lung tumors and to a lesser degree in melanomas.[85,86] The tyrosine kinase and VEGF receptor inhibitor sunitinib decrease tumor burden, increased tumor necrosis and prolonged survival in KL GEMMs.[89] The elevated expression of both HIF1 α and GLUT1 in LKB1 -/- tumors suggests both these proteins may be likely candidates for targeted therapies such as the small molecule inhibitor of GLUT1, STF31, which was recently identified for the treatment of renal cell carcinoma,[87,88] [Figure 2]. The advent of recent phase I/II clinical trials combining metformin and chemotherapies for metastatic breast cancer (Clinical Trial NCT01310231) along with the completion of recent clinical trials testing rapalogs and next generation PI3K/mTOR inhibitors should provide valuable insight into the effectiveness and future of these drugs as realistic therapies for the treatment of LKB1 deficient cancer.[59] Achieving personalized treatments will require defining which oncogenic genotypes sensitize tumors to AMPK activating or mTOR inhibiting drug treatments and is an important goal for future studies.

ACKNOWLEDGMENT

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