HHS Public Access

Author manuscript

Oncogene. Author manuscript; available in PMC 2017 April 21.

Published in final edited form as:

Oncogene. 2017 April 20; 36(16): 2191–2201. doi:10.1038/onc.2016.363.

mTORC1 and mTORC2 in cancer and the tumor microenvironment

Laura C. Kim[1], **Rebecca S. Cook**[1,3], and **Jin Chen**[1,2,3,4,5,#]

[1]Department of Cancer Biology, Vanderbilt University, Nashville, TN 37232

[2]Division of Rheumatology and Immunology, Department of Medicine, Vanderbilt University, Nashville, TN 37232

[3]Vanderbilt-Ingram Cancer Center, Vanderbilt University, Nashville, TN 37232

[4]Department of Cell & Developmental Biology, Vanderbilt University, Nashville, TN 37232

[5]Veterans Affairs Medical Center, Tennessee Valley Healthcare System, Nashville, TN 37212

Abstract

The mammalian target of rapamycin (mTOR) is a crucial signaling node that integrates environmental cues to regulate cell survival, proliferation, and metabolism, and is often deregulated in human cancer. mTOR kinase acts in two functionally distinct complexes, mTOR complex 1 (mTORC1) and 2 (mTORC2), whose activities and substrate specificities are regulated by complex co-factors. Deregulation of this centralized signaling pathway has been associated with a variety of human diseases including diabetes, neurodegeneration, and cancer. While mTORC1 signaling has been extensively studied in cancer, recent discoveries indicate a subset of human cancers harboring amplifications in mTORC2-specific genes as the only actionable genomic alterations, suggesting a distinct role for mTORC2 in cancer as well. This review will summarize recent advances in dissecting the relative contributions of mTORC1 versus mTORC2 in cancer, their role in tumor-associated blood vessels and tumor immunity, and provide an update on mTOR inhibitors.

Introduction

Mammalian Target of Rapamycin (mTOR) is a serine/threonine kinase that was discovered in the early 1990s as the target of the anti-fungal drug rapamycin (1,2). mTOR signaling integrates a variety of environmental and intracellular cues to coordinate a number of cellular processes. The physiological relevance of mTOR signaling is vividly illustrated by the multitude of human diseases that can occur upon its deregulation, including cancer.

Users may view, print, copy, and download text and data-mine the content in such documents, for the purposes of academic research, subject always to the full Conditions of use: http://www.nature.com/authors/editorial_policies/license.html#terms

[#]Correspondence should be addressed to: Jin Chen, M.D., Ph.D., Professor of Medicine, Cancer Biology, and Cell and Developmental Biology, T-3207E, Medical Center North, Vanderbilt University School of Medicine, 1161 21st Avenue South, Nashville, TN 37232, jin.chen@vanderbilt.edu, Telephone 615-343-3819.

Conflicts of Interest: The authors declare no potential conflicts of interest.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Cancer is a disease characterized by its hallmarks (3), including uncontrolled cell proliferation, increased cell survival, evasion of anti-tumor immunity, aberrant angiogenesis, and acquisition of metabolic events unique to cancers. Importantly, activation of mTOR signaling is associated with each of these oncogenic cellular processes, making mTOR a promising target for treating multiple hallmarks of the cancer phenotype.

The mTOR Complexes

Although rapamycin was originally defined as an anti-fungal agent, it was soon realized that rapamycin possesses broad anti-proliferative, cytostatic effects in a wide variety of cells, including cancer cells. Subsequent molecular analyses revealed that rapamycin binds to FKBP12, and in doing so, blocks some (but not all) mTOR activity. In searching for the molecular underpinnings of why rapamycin produced only partial mTOR inhibition, it was discovered that mTOR acts in two functionally distinct complexes (4,5), one that is relatively sensitive to rapamycin (mTOR complex 1, or mTORC1), and one that is relatively rapamycin resistant (mTORC2) (6). Both mTORC1 and mTORC2 harbor several common components: the mTOR kinase, which acts as the central catalytic component, the scaffolding protein mLST8, mTOR regulatory subunit DEPTOR, and the Tti1/Tel2 complex, which is important for mTOR complex assembly and stability. Additionally, each complex harbors distinct subunits (Figure 1) that contribute to substrate specificity, subcellular localization, and complex specific regulation. mTORC1 is defined by its association with Raptor, a scaffolding protein important for mTORC1 assembly, stability, substrate specificity, and regulation, and PRAS40, a factor that blocks mTORC1 activity until growth factor receptor signaling relieves PRAS40-mediated mTORC1 inhibition. The recently solved structure of mTORC1 shows that it acts as a lozenge shaped dimer with the kinase domains coming in close proximity to one another in the center of the structure and Raptor and mLST8 binding on the periphery (7,8).

Rictor and mSin1 are subunits specific to mTORC2. Genetic engineering of cells deficient for Rictor demonstrate that Rictor is required for mTORC2 assembly, stability, substrate identification, and subcellular localization of mTORC2 to the appropriate sites of action (4). mSin1 is also required for subcellular localization of mTORC2 to the plasma membrane (9). Importantly, mSin1 is a key negative regulator of mTORC2 kinase activity, until growth factor receptor-derived signaling through the phosphatidylinositol-3-kinase (PI3K) recruits mSin1/mTORC2 to the plasma membrane, where Sin1-mediated mTORC2 inhibition is relieved. Although the structure of mammalian mTORC2 has yet to be resolved, cross-linking mass spectrometry and electron microscopy have been used to determine the architecture of TORC2 in yeast (10). The structure of TORC2 looks similar to that of TORC1, although TORC2 specific components bind to different locations along the TOR kinase. Since yeast has separate TOR kinases for each complex, solving the structure of mammalian mTORC2 is still an important goal that will lead to further understanding of mTORC2 function.

The differing components and structures of mTORC1 and mTORC2 allow for independent regulation through subcellular localization. For example, active mTORC2 associates closely with the plasma membrane, and has been detected in association with ribosomal membranes

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

(11), where it can interact with its key substrates, the AGC kinases including AKT1-3, serum glucose kinase (SGK) isoforms, and protein kinase C (PKC) family members. In contrast, mTORC1 appears to be affiliated with endosomal and lysosomal membranes, where it interacts with its effectors 4EBP1 and S6K1 (Figure 2).

mTORC1 Signaling

The PI3K pathway is frequently activated in response to oncogenic growth factor receptor signaling. PIK3CA activating mutations, RAS mutations, or PTEN loss result in increased production of the second messenger phosphatidylinositol (3,4,5)-triphosphate (PIP3) (12). Although PIP3 directly recruits and activates mTORC2, PI3K signaling also indirectly activates mTORC1, primarily through AKT. Activation of AKT occurs through phosphorylation at Ser473 mediated by mTORC2, and at T308 mediated by PDK1, another serine-threonine kinase recruited to the plasma membrane by PIP3. Once activated, AKT phosphorylates tuberous sclerosis complex 2 (TSC2), blocking its association with TSC1 (13–15). Since TSC1/2 is a negative regulator of the mTORC1 activator RHEB, AKT-mediated TSC2 phosphorylation allows GTP-loaded RHEB to bind and activate mTORC1 (16,17). AKT also phosphorylates the mTORC1 inhibitor PRAS40, causing PRAS40 to dissociate from Raptor, permitting mTORC1 activation (18–21).

In addition to the PI3K pathway, the Ras-MAPK signaling cascade can activate mTORC1. Similar to AKT-mediated phosphorylation of TSC2, ERK and RSK also phosphorylate TSC2 (22,23), albeit at different residues, to inhibit the TSC1/2 complex and trigger RHEB-mediated activation of mTORC1. Similarly, RSK can also phosphorylate PRAS40 (24), leading to dissociation from Raptor and promoting mTORC1 activation.

Although growth factor signaling through the PI3K/AKT and Ras-MAPK cascades is a key trigger for cellular proliferation, it is important that cells do not proceed with proliferation if the necessary nutrients, energy, and macromolecules are not available to support the high demands of cellular replication. Consistent with this notion, mTORC1 is highly responsive to intracellular ATP, glucose, and certain amino acids, including leucine, arginine and glutamine. Low ATP/high AMP levels activate AMP kinase (AMPK), an indirect mTORC1 inhibitor which functions by promoting TSC1/2 complex formation (25). Thus, AMP accumulation would override growth factor signals and block cellular proliferation in the absence of a sufficient energy supply. Similarly, a lack of amino acids would prevent localization of mTORC1 to lysosomal surfaces where RHEB activates mTORC1, overriding growth factor receptor-derived proliferation signals (26,27), and blocking mTORC1-dependent proliferation in the absence of the needed supply of amino acids. Interestingly, the intracellular location and type of amino acid can be sensed by the cell to determine the mechanism by which mTORC1 lysosomal localization is regulated (28). For example, intralysosomal arginine (29) or cytoplasmic leucine (30–32) activates RAG-GTPases which associate with Raptor directly to localize mTORC1 to lysosomal membranes (33). Meanwhile, cytoplasmic glutamine regulates mTORC1 localization through RAG-independent mechanisms (34,35).

Author Manuscript

Once activated, mTORC1 phosphorylates substrates, including elongation initiation factor (EIF)-4E binding protein 1 (4EBP1) and ribosomal protein S6 kinase 1 (S6K1), two proteins that are key regulators of both cap-dependent and cap-independent translation. Interestingly, increased cap-dependent translation caused by aberrant mTORC1 activation results in increases in cell size (36) and proliferation (37), two common traits of cancer. 4EBP1 and S6K1 bind to eIF-4E and eIF-3, respectively, inhibiting formation of the translation initiating complex. mTORC1-mediated phosphorylation of 4EBP1 and S6K1 liberates their respective binding partners, facilitating preinitiation complex formation (38). S6K1 phosphorylates eIF-4B and S6 ribosomal protein (S6RP), initiating translation (39). S6K1 also plays a key role in translational elongation, phosphorylating eukaryotic elongation factor 2 kinase (eEF2K), allowing eEF2 to continue translational elongation (40). Interestingly, mTORC1 does not affect all transcripts equally. For instance, prostate cancer studies showed that the most common targets of increased translation were those involved in invasion, metastasis, and protein synthesis, highlighting the role of mTORC1 in oncogenic translation (41). Increased translation of protein synthesis genes, consisting mostly of genes involved in ribosomal biogenesis (42–44), is a well-known phenomenon related to mTORC1 hyper-activation. This is a logical target for mTORC1-mediated oncogenic translation since sufficient ribosome levels are required to maintain the increased translation of other genes important for transformation.

mTORC1 in Cancer

Direct evidence for mTORC1 activity in tumorigenesis comes from Tuberous Sclerosis, a disease caused by loss of TSC1 or TSC2, consequently hyper-activating mTORC1, and resulting in widespread but benign tumor formation. The limited progression of these tumors may be due to mTORC1-mediated negative feedback on insulin receptor substrate (IRS)-1, potently downregulating PI3K signaling downstream of most receptor tyrosine kinases (RTKs) (45–47). Also, mTORC1 directly phosphorylates Grb10, an adaptor that directly binds RTKs (48,49), although Grb10 phosphorylation is reported to have the capacity to stimulate and block PI3K activation, perhaps in isoform-specific fashions. Regardless, tuberous sclerosis patients demonstrate that mTORC1 signaling as a single molecular aberration is a potent driver of cellular proliferation. In the context of added genetic and molecular alterations, mTORC1 signaling potentiates the severity of tumor progression through numerous molecular mechanisms.

Transformed cells display metabolic reprogramming, a requirement that may enable cancer to surmount the demands of rapid proliferation. For example, many tumors display aerobic glycolysis, in which glycolysis occurs in the presence of oxygen, perhaps not as a main source of ATP, but rather as a generator of building blocks that can be shunted to alternative anabolic pathways to generate molecules needed for proliferation, including lipids, amino acids, and nucleotides. There is evidence that mTORC1 regulates aerobic glycolysis through increased translation of hypoxia inducible factor (HIF)-1α (50), a transcription factor that drives expression of several glycolytic enzymes (51). mTORC1 upregulates the synthesis of lipids from glycolysis-derived intermediates through phosphorylation of Lipin1 and S6K1, thus activating the transcription factor sterol regulatory element binding factor (SREBP)-1, driving transcription of genes involved in lipogenesis (52,53). Loss of mTORC1-mediated

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

activation of SREBP1 in breast cancer cells blocked lipogenesis, interfering with cellular proliferation and tumor growth (54). Shunting of glycolytic intermediates into nucleotide synthesis is also controlled in part by mTORC1. mTORC1-mediated phosphorylation of S6K1 stimulates both purine and pyrimidine synthesis, which is necessary for cancer cells to rapidly duplicate their DNA (55–57).

These studies would suggest that targeted inhibition of glycolysis would be a feasible approach to blocking cell growth, despite mTORC1 activation. However, phase I clinical trials of the glycolysis inhibitor 2DG yielded disappointing results, with disease progression in the majority of cases, although a few showed stable disease or partial responses (58). Ovarian cancer cells cultured in 2DG to select for glycolysis resistance had upregulated mTORC1 activity as well as increased lipogenesis and nucleotide synthesis (59). These findings suggest that mTORC1-mediated glycolysis may support cancer cells, but that in the absence of glycolysis, other mTORC1-mediated anabolic pathways still support cell proliferation. However, how these tumor cells are supplied with the necessary intermediates for shunting into alternative metabolic pathways, but in the absence of glycolysis, is a question that remains.

Answers may be found in the ability of cells to upregulate macropinocytosis, the process used by cells to obtain macro-nutrients from the extracellular environment. mTORC1 negatively regulates lysosomal degradation of extracellular protein taken up by macropinocytosis. Experiments in which tumor cells were starved of amino acids in culture and in tumors that were poorly vascularized in vivo, demonstrated that mTORC1 inhibition provided a growth advantage, through upregulation of macropinocytosis and catabolism of engulfed proteins (60). The dual roles of mTORC1 in tumor metabolism and growth will need further consideration, particularly in patients treated with mTOR inhibitors.

Aside from regulating cell growth and metabolism, mTORC1 also controls autophagy, an intracellular process that allows orderly degradation and recycling of cellular components. mTORC1 negatively regulates autophagy by phosphorylation of ULK to block initiation of autophagy, VPS34 to block autophagosome formation (61). Autophagy is sometimes considered a tumor suppressor (62), since blockade of autophagy, through deletion of Beclin1 for example, promotes tumor formation (63,64). However, there is abundant evidence to support that autophagy can be harnessed by tumor cells to drive survival under conditions of metabolic duress (65). In this scenario, inhibition of mTORC1 might enhance autophagy, and in doing so, may allow cells to generate nutrients and molecular building blocks to support tumor cell survival to a greater extent than if mTORC1 signaling was left intact.

mTORC2 Signaling

Unlike mTORC1, the upstream regulation of mTORC2 is not well defined, although growth factor stimulation and ribosome association are both known mTORC2 activators (11,66). Importantly, mTORC2 localization at the cell membrane through the mSin1 subunit places mTORC2 in close proximity to its substrates AKT, SGK, and PKC. Thus localization at the plasma membrane is a key aspect of mTORC2 regulation (9). An oncogenic mutation in the

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

PH domain of mSin1 that blocks mSIN1-mediated mTOR inhibition leading to constitutive mTORC2-AKT signaling has been identified in an ovarian cancer patient (9). Direct phosphorylation of mSin1 at T86 by AKT may also regulate mTORC2, leading to a positive feedback loop that sustains mTORC2-AKT signaling, while mSin1 phosphorylation by S6K1 at this same site may inhibit mTORC2 activity as a feedback mechanism downstream of mTORC1 (67–69). Recent studies have identified another function for mSin1 in mTORC2 regulation. The PH domain of mSin1 can also bind to phosphorylated cytoplasmic Rb through to inhibit mTORC2 complex formation and reduce AKT signaling (70). Regulation of mTORC2 by mSin1 may be cell-type and/or context dependent. Clarification of these feedback mechanisms will be important for complete understanding of mTORC2 activation and signaling in cancer.

AKT, a key substrate of mTORC2, is among the most commonly hyper-activated proteins in cancer. AKT integrates signals from PI3K/mTORC2 and from PI3K/PDK1 to promote cell growth and survival. Like mTORC2, AKT localization to the plasma membrane is regulated by PIP3. In PTEN null prostate cancer, loss of mTORC2 activity inhibits tumorigenesis, illustrating the importance of mTORC2 signaling downstream of PIP3 (71). Interestingly, PTEN null glioma patients exhibit mTORC2-mediated chemotherapy resistance in an AKT independent manner (72), suggesting that inhibition of mTORC2 may be useful in treatment of patients with PTEN or PI3K mutations. mTORC2 also regulates cancer cells' preferential use of glycolysis for energy production through the AKT-independent acetylation of FoxO1/3 (73), demonstrating a mTORC2-mediated role in cancer metabolism. In addition to its activation by mTORC2, AKT activates mTORC1 signaling (13,21), adding another layer of complexity to this signaling pathway.

mTORC2 also phosphorylates SGK and PKC family members. Activation of SGK3 is implicated in cancer particularly because of its ability to reinforce PI3K signaling through INPP4B (74). Importantly, SGK1 promotes resistance to chemotherapy (75) and AKT inhibitors (76). Substrates of SGK include both NDRG1 and FoxO family transcription factors, two factors that are not growth promoting under oxygen and/or nutrient replete conditions, but which can promote survival in response to oxygen or nutrient deprivation, or in response to PI3K inhibition (75,77). NDRG1 is a potent suppressor of tumor cell invasion and metastasis, and is degraded in response to SGK-mediated phosphorylation.

PKC, which exists in several isoforms, is also known to be involved in tumorigenesis, although the exact role of each isoform has yet to be defined. Studies have shown that each isoform may work in a cell-type specific manner. In the mouse mammary gland, genetic disruption of Rictor blocked mTORC2-dependent ductal branching, and reduced motility, invasion, and survival of mammary epithelial cells. Importantly this was rescued upon reactivation of PKCα and its downstream effector, Rac1 (78). Although studied in the developmental setting, all of these phenotypes are known to be important in breast cancer metastasis, suggesting a role for the mTORC2/PKCα signaling axis in breast cancer as well.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

mTORC2 in Cancer

While mTORC1 is extensively studied in cancer, recent reports also demonstrate a distinct role for mTORC2 in prostate, breast, and lung cancer, glioblastoma, and T-cell acute lymphoblastic leukemia (T-ALL) (71,79). Amplification of *RICTOR* was observed in non-small cell lung cancer patients (80), breast cancer (81), and in residual disease of triple-negative breast cancers treated with neoadjuvant chemotherapy (82), reinforcing the importance of mTORC2 signaling in cancer and as a potential target for inhibition. Rictor overexpression was also previously noted in gliomas, in which about 70% of patients have increased AKT activity (83). In HER2-positive breast cancer, enriched Rictor expression leads to hyper-activation of AKT and tumor progression. Knockdown of Rictor (but not Raptor) or treatment with mTORC1/2 dual kinase inhibitors (but not mTORC1-specific rapalogs) decreased AKT-mediated tumor cell survival and increased therapeutic tumor cell killing in cells treated with the HER2/EGFR tyrosine kinase inhibitor, lapatinib (81). Collectively, these data suggest a distinct role for mTORC2 in cancer.

mTOR Inhibitors for Cancer Therapy

Rapamycin was the original mTOR inhibitor, which allosterically inhibits mTORC1 (Figure 1), but not mTORC2. The yeast structure of TORC2 suggests the Rictor analog Avo3 may block FKBP12-rapamycin complex binding to the mTOR kinase, granting rapamycin insensitivity, although the lack of mammalian mTORC2 structure prevents confirmation of this mechanism in humans (10). Interestingly, prolonged treatment with rapamycin inhibits mTORC2 in certain cell types, suggesting a cell-type specific mechanism of regulation of mTORC2 (84). Rapamycin analogs ("rapalogs") have been developed (Table 1) with enhanced pharmacokinetic properties for more effective treatment of patients (85). The first rapalog, temsirolimus, was approved by the FDA in 2007 after it was shown to be effective in treating advanced renal cell carcinoma (86). Since then, everolimus, another rapalog, has been approved for treatment of several other cancers including breast, pancreatic, lung, and subependymal giant cell astrocytoma. Rapalogs most often cause disease stabilization rather than regression, consistent with the idea that mTORC1 is a driver of cellular proliferation, but not cell survival.

There are several potential reasons for the limited efficacy of rapalogs in treating cancer. First, inhibition of mTORC1 action on its substrates is incomplete. Inhibition of mTORC1 by rapamycin completely blocks the phosphorylation of S6K1, but phosphorylation of 4EBP1 is often only modestly inhibited (87). Since 4EBP1 regulates cap-dependent translation, it is possible that 4EBP1 is still able to translate proteins important in tumorigenesis. Additionally, inhibition of mTORC1 will release mTORC1-mediated restraints on PI3K/mTORC2/AKT signaling, resulting in resurgent AKT signaling, increased growth, and heightened cell survival (88,89). mTORC1 inhibition may also cause increased cell proliferation within vascularly compromised tumor regions due to elevated micropinocytosis (60) of extracellular proteins or increased cell survival through enhanced autophagy (65).

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Recent advances have been realized in ATP-competitive mTOR kinase inhibitors (TOR-KIs), which block mTOR catalytic activity, whether embedded within mTORC1 or mTORC2 (Figure 1) (87). Preclinical testing of these inhibitors has shown complete 4EBP1 inhibition, with sustained repression of mTORC2-mediated AKT phosphorylation, resulting in superior tumor cell killing and growth inhibition as compared to rapalogs (89). mTOR kinase inhibitors are currently in Phase II clinical trials, after promising results in Phase I trials (Table 2) (91,92). While partial responses and stable disease have been reported, toxicity and adverse side effects are still a concern.

Pre-clinical studies have identified mutations in both the kinase domain and FRB domain of mTOR that prevent binding of either rapalogs or ATP-competitive mTOR kinase inhibitors, leading to loss of efficacy and eventual resistance. Most recently, a third-generation mTOR inhibitor has been developed to overcome resistance to currently available inhibitors. This third-generation inhibitor has a bivalent structure consisting of a rapamycin-FRB binding element linked to a TOR-KI (93), so that when at least one half the ligand binds, the other half is in close proximity to the second binding site, overcoming point mutations that prevent binding of either drug alone. Even with development of third-generation mTOR inhibitors, concerns about toxicity remain. Additionally, mTORC1 inhibition, even within the context of mTORC2 inhibition, may promote tumor cell proliferation under nutrient-stressed conditions, as evidenced by the mutant KRAS pancreatic ductal carcinoma tumor model (60). Therefore, it will be important to investigate the impact of TOR-KIs on macropinocytosis and autophagy in distinct tumor types under both nutrient replete and deprived conditions.

Small molecular weight kinase inhibitors capable of simultaneous blockade of mTOR and PI3K have also been developed (Table 2). As expected, these inhibitors overcome the limitations of rapalogs and inhibit mTORC2-independent activation of AKT, while providing superior blockade of resurgent PI3K activity. Unfortunately, Phase I clinical trials using PI3K/mTOR dual kinase inhibitors revealed significant dose-limiting on-target toxicities (94,95), consistent with the important roles these enzymes fulfill in homeostasis of healthy tissues and systemic metabolism.

There is significant interest in developing mTORC2 specific inhibitors that will leave the activities of mTORC1 intact. Selectively targeting the mTORC2 branch may avoid feedback loop inhibition caused by rapalogs, and may be particularly effective in vascularly compromised tumors under metabolic stress. As an added and important benefit, toxicities related to mTORC1 inhibition, including lesions within the oral mucosa, rash, and immune suppression, may also be reduced. Notably, genomic aberrations in Rictor and mSin1 have been identified in several tumor types (9,80,81,83) and patients with these genetic alterations may benefit from an mTORC2 specific inhibitor.

mTOR in Vasculature

In addition to its essential role in tumor cells, mTOR signaling is critical in the tumor microenvironment (TME) (Table 3). For example, mTOR is key for tumor angiogenesis (96,97), a well-studied hallmark of cancer. In response to oxygen and/or nutrient

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

deprivation, tumor cells secrete factors that recruit new vessel formation to support the growing tumor. Blockade of tumor angiogenesis would effectively limit tumor growth. Additionally, tumor vessels provide a route for tumor cells to disseminate to distant sites; as such, blockade of tumor angiogenesis could be harnessed to prevent tumor metastasis.

Hypoxia in the tumor stimulates angiogenesis via HIF transcription factors. In some cases, oncogenic mTOR signaling can actively promote cap-dependent translation of HIF-1α (98,99). HIF-1α activates expression of proangiogenic factors that are secreted by the tumor cell, including vascular endothelial growth factor (VEGF). VEGF binds to the VEGF receptors on the surface of vascular endothelial cells, to promote angiogenesis. Interestingly, loss of the mTORC1 negative regulator TSC1 from vascular endothelial cells drives proliferative lesions resembling lymphangiosarcoma, suggesting that mTORC1 is a dominant driver of endothelial cell proliferation(100), although evidence suggests mTORC2 could also play a role in endothelial cell proliferation through downstream effector PKCα (101). TSC1-deficient lymphangiosarcoma formation was suppressed not only by rapamycin, but also was by inhibitors of VEGF, defining a mTORC1-VEGF feed-forward loop in the angiogenic process that drives endothelial cell proliferation, survival, and vascular assembly (100). Rapalogs have been successful in treating highly vascularized tumors like Kaposi’s sarcoma and renal cancer.

Interest in mTORC2 activity in the tumor vasculature was initiated with the observation that mTORC2 loss from endothelial cells causes deficiencies in physiological vascular development (102). Importantly, mTORC2 signaling has been implicated in sprouting angiogenesis stimulated by VEGF (103) and CXCL12 (104), both factors secreted by tumor cells to promote a more favorable microenvironment. Downstream of mTORC2, aberrant AKT signaling within the vascular endothelium promotes the tortuous and leaky vascular structures often associated with tumors (105). AKT has also been shown to primarily regulate vascular endothelial cell assembly (101). FoxO1, a substrate of the mTORC2 effectors AKT and SGK, has also been implicated in endothelial cell viability (106), growth (107), and metabolism (108). Since AKT can also activate mTORC1, it is possible that mTORC2 may have regulatory functions in the autocrine VEGF signaling within the vascular endothelium (100,109,110).

The use of mTOR inhibitors in treating cancers has provided insight into the effects of these inhibitors on the tumor vasculature. Everolimus, like VEGF inhibitors, decreased the tumor vasculature associated with a variety of solid tumor cell lines. While VEGF inhibitors were more potent at blocking formation of new vessels, everolimus was effective at reducing the viability of existing vessels (111). Consistent with the ability of everolimus to impair the integrity of existing tumor vasculature, radiation therapy caused excess damage to vascular endothelial cells upon mTOR kinase inhibition (112,113).

mTOR in Tumor Immunity

Along with the tumor vasculature, the immune system is another facet of the TME that supports tumor initiation, progression, and metastasis, as tumor cells must actively evade immune surveillance to prevent eradication by the host. Tumors often express

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

immunosuppressive 'checkpoint' markers such as CTLA-4 and PD-L1/PD-L2, that anergize CD8+ T-cells that would otherwise mount a cytotoxic attack (114). Inhibiting these immune checkpoints allows cytotoxic CD8+ T-cell activation and anti-tumor immune responses. Anti-PD-1/PD-L1 therapy has been extraordinarily successful in the treatment of melanoma patients and is currently in clinical trials for many other types of cancer.

A recent study in non-small cell lung cancer cells demonstrated that hyperactivated AKT-mTOR signaling directly increases PD-L1 expression (115). Interestingly, melanoma tumors grown in immunocompromised mice still respond to anti-PD1 therapy, suggesting a tumor intrinsic role for these immune checkpoint molecules (116). Further investigation showed that PD-1:PD-L1 interaction on tumor cells signals through mTORC1 to promote tumor cell proliferation. Together, these data suggest combination of checkpoint inhibitors and mTOR inhibition may be more efficacious than either therapy alone.

mTOR signaling within the immune cells themselves also deserves scrutiny, since rapamycin was first used in the clinic as an immune suppressant in organ transplant patients. The role of mTOR signaling in determining the fates of helper T cells has been relatively well defined. mTORC1 activity is predominately associated with Th1 differentiation and anti-tumor immunity (117). Inhibition of mTORC1 using rapalogs or rapamycin encourages engraftment in organ transplant patients through inhibition of cytotoxic immunity, particularly through expansion of CD4+regulatory T cells (T_{reg}) (118). In contrast to mTORC1, mTORC2 activity is often associated with immune suppressive Th2 phenotypes (117,119). This is regulated by mTORC2 downstream effector, SGK, which when depleted, limits differentiation of Th2 CD4+ T-cells (120).

In addition to CD4+ T-cells, mTOR signaling also regulates CD8+ T-cell effector function and differentiation, both important processes for mounting an immune response towards tumor cells. The role of mTORC1 as a regulator of CD8+ T-cell effector function has been well-defined by genetic deletion of the mTORC1 suppressor TSC2 in T-lymphocytes which resulted in mTORC1 upregulation and profound CD8+ T-cell effector function (121). Conversely, the role of mTOR signaling in establishing CD8+ memory T-cells has yet to be clearly defined. Rapamycin treatment reduced mTORC1 activity and increased CD8+ memory T-cell formation (122). Contrarily, other reports suggest that memory T-cell formation is predominately regulated by the mTORC2 pathway, as demonstrated by an increase in memory T-cell formation upon genetic deletion of Rictor in T-lymphocytes (121,123). This discrepancy may be explained by the ability for rapamycin to inhibit mTORC2 in certain cell types, which would confirm the role of mTORC2 as the predominant regulator of memory T-cell differentiation.

While the role of T-cells is currently most well-studied in terms of mTOR signaling and tumor immunity, there is also evidence that mTOR signaling plays a role in other immune cells that could be important in the tumor microenvironment. TSC2 deletion in myeloid lineages increased mTORC1 signaling and blocked differentiation of macrophages towards an M2 phenotype, the phenotype most closely correlating to pro-malignant and immune suppressive tumor-associated macrophages (TAMs) (124). Although the roles played by B cells in tumor immunity is less understood, it is known B-cells can promote inflammation

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

and carcinogenesis, as well as regulate anti-tumor T-cell responses. mTOR signaling is implicated in physiological B cell maturation, survival and proliferation, suggesting a possible role for mTOR in antibody-mediated regulation of tumor immunity (125).

Conclusions and Future Directions

Increasing investigations of mTOR signaling in cancer cells and throughout the complex TME has provided the platforms from which new studies will reveal a more refined understanding of mTOR within each tumor compartment, the distinct and intertwining roles of mTORC1 and mTORC2, and how this knowledge can be applied towards novel therapeutic strategies that will safely and effectively eradicate cancers. Although rapalogs and mTOR kinase inhibitors have been used to treat patients with mTOR-dependent cancers, they have also exposed deficiencies in our knowledge of mTOR signaling and its role in cancer. As more patients are treated with these drugs, it will be important to not only monitor changes in tumor size and progression, but changes to the vasculature and immune system as well. In addition, mTORC2 specific inhibitors might be explored for use both in the laboratory and clinical settings. Preclinical studies of mTOR kinase inhibitors exposed new mechanisms of action for mTORC1. We anticipate similarly serendipitous findings as studies place increasing focus on mTORC2.

Acknowledgments

Funding: This work was supported by the Department of Veterans Affairs through a VA Merit Award and a Research Career Scientist Award (J. Chen), NIH grants R01 CA95004 and R01 CA177681 (J. Chen), and T-32 CA009592 (L. Kim).

We thank Deanna Edwards for careful review of the manuscript. This work was supported by the Department of Veterans Affairs through a VA Merit Award and a Research Career Scientist Award (J. Chen), NIH grants R01 CA95004 and R01 CA177681 (J. Chen), and T-32 CA009592 (L. Kim).

References

1. Brown EJ, Albers MW, Bum Shin T, ichikawa K, Keith CT, Lane WS, et al. A mammalian protein targeted by G1-arresting rapamycin–receptor complex. Nature. 1994 Jun 30; 369(6483):756–8. [PubMed: 8008069]
2. Sabatini DM, Erdjument-Bromage H, Lui M, Tempst P, Snyder SH. RAFT1: A mammalian protein that binds to FKBP12 in a rapamycin-dependent fashion and is homologous to yeast TORs. Cell. 1994 Jul; 78(1):35–43. [PubMed: 7518356]
3. Hanahan D, Weinberg RA. Hallmarks of cancer: the next generation. Cell. 2011 Mar 4; 144(5):646–74. [PubMed: 21376230]
4. Sarbassov DD, Ali SM, Kim D-H, Guertin DA, Latek RR, Erdjument-Bromage H, et al. Rictor, a novel binding partner of mTOR, defines a rapamycin-insensitive and raptor-independent pathway that regulates the cytoskeleton. Curr Biol. 2004 Jul 27; 14(14):1296–302. [PubMed: 15268862]
5. Jacinto E, Loewith R, Schmidt A, Lin S, Rüegg MA, Hall A, et al. Mammalian TOR complex 2 controls the actin cytoskeleton and is rapamycin insensitive. Nat Cell Biol. 2004 Nov; 6(11):1122–8. [PubMed: 15467718]
6. Laplante M, Sabatini DM. mTOR signaling in growth control and disease. Cell. 2012 Apr 13; 149(2):274–93. [PubMed: 22500797]
7. Yang H, Rudge DG, Koos JD, Vaidialingam B, Yang HJ, Pavletich NP. mTOR kinase structure, mechanism and regulation. Nature. 2013 May 9; 497(7448):217–23. [PubMed: 23636326]

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

8. Aylett CHS, Sauer E, Imseng S, Boehringer D, Hall MN, Ban N, et al. Architecture of human mTOR complex 1. Science (80-) American Association for the Advancement of Science. 2015 Dec 17; 351(6268):48–52.
9. Liu P, Gan W, Chin YR, Ogura K, Guo J, Zhang J, et al. PtdIns(3,4,5)P3-Dependent Activation of the mTORC2 Kinase Complex. Cancer Discov. 2015 Aug 20.
10. Gaubitz C, Oliveira TM, Prouteau M, Leitner A, Karuppasamy M, Konstantinidou G, et al. Molecular Basis of the Rapamycin Insensitivity of Target Of Rapamycin Complex 2. Mol Cell. 2015 Jun 18; 58(6):977–88. [PubMed: 26028537]
11. Zinzalla V, Stracka D, Oppliger W, Hall MN. Activation of mTORC2 by association with the ribosome. Cell. 2011 Mar 4; 144(5):757–68. [PubMed: 21376236]
12. Thorpe LM, Yuzugullu H, Zhao JJ. PI3K in cancer: divergent roles of isoforms, modes of activation and therapeutic targeting. Nat Rev Cancer Nature Publishing Group. 2014 Dec 23; 15(1):7–24.
13. Inoki K, Li Y, Zhu T, Wu J, Guan K-L. TSC2 is phosphorylated and inhibited by Akt and suppresses mTOR signalling. Nat Cell Biol Nature Publishing Group. 2002 Sep 12; 4(9):648–57.
14. Manning BD, Tee AR, Logsdon MN, Blenis J, Cantley LC. Identification of the Tuberous Sclerosis Complex-2 Tumor Suppressor Gene Product Tuberin as a Target of the Phosphoinositide 3-Kinase/Akt Pathway. Mol Cell. 2002; 10(1):151–62. [PubMed: 12150915]
15. Potter CJ, Pedraza LG, Xu T. Akt regulates growth by directly phosphorylating Tsc2. Nat Cell Biol. 2002 Sep; 4(9):658–65. [PubMed: 12172554]
16. Inoki K, Li Y, Xu T, Guan K-L. Rheb GTPase is a direct target of TSC2 GAP activity and regulates mTOR signaling. Genes Dev. 2003 Aug 1; 17(15):1829–34. [PubMed: 12869586]
17. Tee AR, Manning BD, Roux PP, Cantley LC, Blenis J. Tuberous Sclerosis Complex Gene Products, Tuberin and Hamartin, Control mTOR Signaling by Acting as a GTPase- Activating Protein Complex toward Rheb. Curr Biol. 2003; 13(15):1259–68. [PubMed: 12906785]
18. Sancak Y, Thoreen CC, Peterson TR, Lindquist RA, Kang SA, Spooner E, et al. PRAS40 Is an Insulin-Regulated Inhibitor of the mTORC1 Protein Kinase. Mol Cell. 2007; 25(6):903–15. [PubMed: 17386266]
19. Thedieck K, Polak P, Kim ML, Molle KD, Cohen A, Jenö P, et al. PRAS40 and PRR5-like protein are new mTOR interactors that regulate apoptosis. PLoS One. 2007; 2(11):e1217. [PubMed: 18030348]
20. Vander Haar E, Lee S-I, Bandhakavi S, Griffin TJ, Kim D-H. Insulin signalling to mTOR mediated by the Akt/PKB substrate PRAS40. Nat Cell Biol. 2007 Mar; 9(3):316–23. [PubMed: 17277771]
21. Wang L, Harris TE, Roth RA, Lawrence JC. PRAS40 regulates mTORC1 kinase activity by functioning as a direct inhibitor of substrate binding. J Biol Chem. 2007 Jul 6; 282(27):20036–44. [PubMed: 17510057]
22. Ma L, Chen Z, Erdjument-Bromage H, Tempst P, Pandolfi PP. Phosphorylation and Functional Inactivation of TSC2 by Erk: Implications for Tuberous Sclerosisand Cancer Pathogenesis. Cell. 2005; 121(2):179–93. [PubMed: 15851026]
23. Roux PP, Ballif BA, Anjum R, Gygi SP, Blenis J. Tumor-promoting phorbol esters and activated Ras inactivate the tuberous sclerosis tumor suppressor complex via p90 ribosomal S6 kinase. Proc Natl Acad Sci U S A. 2004 Sep 14; 101(37):13489–94. [PubMed: 15342917]
24. Carrière A, Cargnello M, Julien L-A, Gao H, Bonneil É, Thibault P, et al. Oncogenic MAPK Signaling Stimulates mTORC1 Activity by Promoting RSK-Mediated Raptor Phosphorylation. Curr Biol. 2008; 18(17):1269–77. [PubMed: 18722121]
25. Inoki K, Zhu T, Guan K-L. TSC2 Mediates Cellular Energy Response to Control Cell Growth and Survival. Cell. 2003; 115(5):577–90. [PubMed: 14651849]
26. Blommaart EF, Luiken JJ, Blommaart PJ, van Woerkom GM, Meijer AJ. Phosphorylation of ribosomal protein S6 is inhibitory for autophagy in isolated rat hepatocytes. J Biol Chem. 1995 Feb 3; 270(5):2320–6. [PubMed: 7836465]
27. Hara K, Yonezawa K, Weng QP, Kozlowski MT, Belham C, Avruch J. Amino acid sufficiency and mTOR regulate p70 S6 kinase and eIF-4E BP1 through a common effector mechanism. J Biol Chem. 1998 Jun 5; 273(23):14484–94. [PubMed: 9603962]

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

28. Shimobayashi M, Hall MN. Multiple amino acid sensing inputs to mTORC1. Cell Res Nature Publishing Group. 2016 Jan 11; 26(1):7–20.
29. Wang S, Tsun Z-Y, Wolfson RL, Shen K, Wyant GA, Plovanich ME, et al. Metabolism. Lysosomal amino acid transporter SLC38A9 signals arginine sufficiency to mTORC1. Science American Association for the Advancement of Science. 2015 Jan 9; 347(6218):188–94.
30. Han JM, Jeong SJ, Park MC, Kim G, Kwon NH, Kim HK, et al. Leucyl-tRNA Synthetase Is an Intracellular Leucine Sensor for the mTORC1-Signaling Pathway. Cell. 2012; 149(2):410–24. [PubMed: 22424946]
31. Bonfils G, Jaquenoud M, Bontron S, Ostrowicz C, Ungermann C, De Virgilio C. Leucyl- tRNA Synthetase Controls TORC1 via the EGO Complex. Molecular Cell. 2012; 46
32. Wolfson RL, Chantranupong L, Saxton RA, Shen K, Scaria SM, Cantor JR, et al. Sestrin2 is a leucine sensor for the mTORC1 pathway. Science (80-) American Association for the Advancement of Science. 2015 Oct 8; 351(6268):43–8.
33. Sancak Y, Peterson TR, Shaul YD, Lindquist RA, Thoreen CC, Bar-Peled L, et al. The Rag GTPases bind raptor and mediate amino acid signaling to mTORC1. Science. 2008 Jun 13; 320(5882):1496–501. [PubMed: 18497260]
34. Stracka D, Jozefczuk S, Rudroff F, Sauer U, Hall MN. Nitrogen source activates TOR (target of rapamycin) complex 1 via glutamine and independently of Gtr/Rag proteins. J Biol Chem. 2014 Sep 5; 289(36):25010–20. [PubMed: 25063813]
35. Jewell JL, Kim YC, Russell RC, Yu F-X, Park HW, Plouffe SW, et al. Metabolism. Differential regulation of mTORC1 by leucine and glutamine. Science. 2015 Jan 9; 347(6218):194–8. [PubMed: 25567907]
36. Fingar DC, Salama S, Tsou C, Harlow E, Blenis J. Mammalian cell size is controlled by mTOR and its downstream targets S6K1 and 4EBP1/eIF4E. Genes Dev. 2002 Jun 15; 16(12):1472–87. [PubMed: 12080086]
37. Dowling RJO, Topisirovic I, Alain T, Bidinosti M, Fonseca BD, Petroulakis E, et al. mTORC1-mediated cell proliferation, but not cell growth, controlled by the 4E-BPs. Science. 2010 May 28; 328(5982):1172–6. [PubMed: 20508131]
38. Ma XM, Blenis J. Molecular mechanisms of mTOR-mediated translational control. Nat Rev Mol Cell Biol Nature Publishing Group. 2009 May; 10(5):307–18.
39. Holz MK, Ballif BA, Gygi SP, Blenis J. mTOR and S6K1 mediate assembly of the translation preinitiation complex through dynamic protein interchange and ordered phosphorylation events. Cell Elsevier. 2005 Nov 18; 123(4):569–80.
40. Browne GJ, Proud CG. A novel mTOR-regulated phosphorylation site in elongation factor 2 kinase modulates the activity of the kinase and its binding to calmodulin. Mol Cell Biol. 2004 Apr; 24(7): 2986–97. [PubMed: 15024086]
41. Hsieh AC, Liu Y, Edlind MP, Ingolia NT, Janes MR, Sher A, et al. The translational landscape of mTOR signalling steers cancer initiation and metastasis. Nature. Nature Publishing Group, a division of Macmillan Publishers Limited. All Rights Reserved. 2012 May 3; 485(7396):55–61.
42. Mayer C, Zhao J, Yuan X, Grummt I. mTOR-dependent activation of the transcription factor TIF-IA links rRNA synthesis to nutrient availability. Genes Dev. 2004 Feb 15; 18(4):423–34. [PubMed: 15004009]
43. Kantidakis T, Ramsbottom BA, Birch JL, Dowding SN, White RJ. mTOR associates with TFIIIC, is found at tRNA and 5S rRNA genes, and targets their repressor Maf1. Proc Natl Acad Sci U S A. 2010 Jun 29; 107(26):11823–8. [PubMed: 20543138]
44. Shor B, Wu J, Shakey Q, Toral-Barza L, Shi C, Follettie M, et al. Requirement of the mTOR kinase for the regulation of Maf1 phosphorylation and control of RNA polymerase III-dependent transcription in cancer cells. J Biol Chem. 2010 May 14; 285(20):15380–92. [PubMed: 20233713]
45. Harrington LS, Findlay GM, Gray A, Tolkacheva T, Wigfield S, Rebholz H, et al. The TSC1-2 tumor suppressor controls insulin-PI3K signaling via regulation of IRS proteins. J Cell Biol. 2004 Jul 19; 166(2):213–23. [PubMed: 15249583]
46. Um SH, Frigerio F, Watanabe M, Picard F, Joaquin M, Sticker M, et al. Absence of S6K1 protects against age- and diet-induced obesity while enhancing insulin sensitivity. Nature. 2004 Oct 9; 431(7005):200–5. [PubMed: 15306821]

Author Manuscript

47. Shah OJ, Wang Z, Hunter T. Inappropriate activation of the TSC/Rheb/mTOR/S6K cassette induces IRS1/2 depletion, insulin resistance, and cell survival deficiencies. Curr Biol. 2004 Sep 21; 14(18):1650–6. [PubMed: 15380067]
48. Hsu PP, Kang SA, Rameseder J, Zhang Y, Ottina KA, Lim D, et al. The mTOR-regulated phosphoproteome reveals a mechanism of mTORC1-mediated inhibition of growth factor signaling. Science American Association for the Advancement of Science. 2011 Jun 10; 332(6035):1317–22.
49. Yu Y, Yoon S-O, Poulogiannis G, Yang Q, Ma XM, Villén J, et al. Phosphoproteomic analysis identifies Grb10 as an mTORC1 substrate that negatively regulates insulin signaling. Science American Association for the Advancement of Science. 2011 Jun 10; 332(6035):1322–6.
50. Majumder PK, Febbo PG, Bikoff R, Berger R, Xue Q, McMahon LM, et al. mTOR inhibition reverses Akt-dependent prostate intraepithelial neoplasia through regulation of apoptotic and HIF-1-dependent pathways. Nat Med. 2004 Jun; 10(6):594–601. [PubMed: 15156201]
51. Düvel K, Yecies JL, Menon S, Raman P, Lipovsky AI, Souza AL, et al. Activation of a metabolic gene regulatory network downstream of mTOR complex 1. Mol Cell. 2010 Jul 30; 39(2):171–83. [PubMed: 20670887]
52. Peterson TR, Sengupta SS, Harris TE, Carmack AE, Kang SA, Balderas E, et al. mTOR complex 1 regulates lipin 1 localization to control the SREBP pathway. Cell. 2011 Aug 5; 146(3):408–20. [PubMed: 21816276]
53. Porstmann T, Santos CR, Griffiths B, Cully M, Wu M, Leevers S, et al. SREBP activity is regulated by mTORC1 and contributes to Akt-dependent cell growth. Cell Metab. 2008 Sep; 8(3):224–36. [PubMed: 18762023]
54. Ricoult SJH, Yecies JL, Ben-Sahra I, Manning BD. Oncogenic PI3K and K-Ras stimulate de novo lipid synthesis through mTORC1 and SREBP. Oncogene. 2016 Mar 10; 35(10):1250–60. [PubMed: 26028026]
55. Ben-Sahra I, Howell JJ, Asara JM, Manning BD. Stimulation of de novo pyrimidine synthesis by growth signaling through mTOR and S6K1. Science. 2013 Mar 15; 339(6125):1323–8. [PubMed: 23429703]
56. Robitaille AM, Christen S, Shimobayashi M, Cornu M, Fava LL, Moes S, et al. Quantitative phosphoproteomics reveal mTORC1 activates de novo pyrimidine synthesis. Science American Association for the Advancement of Science. 2013 Mar 15; 339(6125):1320–3.
57. Ben-Sahra I, Hoxhaj G, Ricoult SJH, Asara JM, Manning BD. mTORC1 induces purine synthesis through control of the mitochondrial tetrahydrofolate cycle. Science (80-) American Association for the Advancement of Science. 2016 Feb 11; 351(6274):728–33.
58. Raez LE, Papadopoulos K, Ricart AD, Chiorean EG, Dipaola RS, Stein MN, et al. A phase I dose-escalation trial of 2-deoxy-D-glucose alone or combined with docetaxel in patients with advanced solid tumors. Cancer Chemother Pharmacol. 2013 Feb; 71(2):523–30. [PubMed: 23228990]
59. Pusapati RV, Daemen A, Wilson C, Sandoval W, Gao M, Haley B, et al. mTORC1-Dependent Metabolic Reprogramming Underlies Escape from Glycolysis Addiction in Cancer Cells. Cancer Cell Elsevier. 2016 Mar 11; 29(4):548–62.
60. Palm W, Park Y, Wright K, Pavlova NN, Tuveson DA, Thompson CB. The Utilization of Extracellular Proteins as Nutrients Is Suppressed by mTORC1. Cell. 2015 Jul 1; 162(2):259–70. [PubMed: 26144316]
61. Kim YC, Guan K-L. mTOR: a pharmacologic target for autophagy regulation. J Clin Invest American Society for Clinical Investigation. 2015 Jan 2; 125(1):25–32.
62. White E. The role for autophagy in cancer. J Clin Invest American Society for Clinical Investigation. 2015 Jan 2; 125(1):42–6.
63. Yue Z, Jin S, Yang C, Levine AJ, Heintz N. Beclin 1, an autophagy gene essential for early embryonic development, is a haploinsufficient tumor suppressor. Proc Natl Acad Sci U S A. 2003 Dec 9; 100(25):15077–82. [PubMed: 14657337]
64. Qu X, Yu J, Bhagat G, Furuya N, Hibshoosh H, Troxel A, et al. Promotion of tumorigenesis by heterozygous disruption of the beclin 1 autophagy gene. J Clin Invest. 2003 Dec; 112(12):1809–20. [PubMed: 14638851]

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

65. Degenhardt K, Mathew R, Beaudoin B, Bray K, Anderson D, Chen G, et al. Autophagy promotes tumor cell survival and restricts necrosis, inflammation, and tumorigenesis. Cancer Cell. 2006 Jul; 10(1):51–64. [PubMed: 16843265]
66. Sarbassov DD, Guertin DA, Ali SM, Sabatini DM. Phosphorylation and regulation of Akt/PKB by the rictor-mTOR complex. Science American Association for the Advancement of Science. 2005 Feb 18; 307(5712):1098–101.
67. Humphrey SJ, Yang G, Yang P, Fazakerley DJ, Stöckli J, Yang JY, et al. Dynamic adipocyte phosphoproteome reveals that Akt directly regulates mTORC2. Cell Metab. 2013 Jun 4; 17(6): 1009–20. [PubMed: 23684622]
68. Yang G, Murashige DS, Humphrey SJ, James DE. A Positive Feedback Loop between Akt and mTORC2 via SIN1 Phosphorylation. Cell Rep. 2015 Jul 29; 12(6):937–43. [PubMed: 26235620]
69. Liu P, Gan W, Inuzuka H, Lazorchak AS, Gao D, Arojo O, et al. Sin1 phosphorylation impairs mTORC2 complex integrity and inhibits downstream Akt signalling to suppress tumorigenesis. Nat Cell Biol. Nature Publishing Group, a division of Macmillan Publishers Limited. All Rights Reserved. 2013 Nov; 15(11):1340–50.
70. Zhang J, Xu K, Liu P, Geng Y, Wang B, Gan W, et al. Inhibition of Rb Phosphorylation Leads to mTORC2-Mediated Activation of Akt. Mol Cell. 2016 May 26.
71. Guertin DA, Stevens DM, Saitoh M, Kinkel S, Crosby K, Sheen J-H, et al. mTOR complex 2 is required for the development of prostate cancer induced by Pten loss in mice. Cancer Cell. 2009 Feb 3; 15(2):148–59. [PubMed: 19185849]
72. Tanaka K, Babic I, Nathanson D, Akhavan D, Guo D, Gini B, et al. Oncogenic EGFR signaling activates an mTORC2-NF-κB pathway that promotes chemotherapy resistance. Cancer Discov American Association for Cancer Research. 2011 Nov 1; 1(6):524–38.
73. Masui K, Tanaka K, Akhavan D, Babic I, Gini B, Matsutani T, et al. mTOR complex 2 controls glycolytic metabolism in glioblastoma through FoxO acetylation and upregulation of c-Myc. Cell Metab. 2013 Nov 5; 18(5):726–39. [PubMed: 24140020]
74. Gasser JA, Inuzuka H, Lau AW, Wei W, Beroukhim R, Toker A. SGK3 mediates INPP4B-dependent PI3K signaling in breast cancer. Mol Cell. 2014 Nov 20; 56(4):595–607. [PubMed: 25458846]
75. Weiler M, Blaes J, Pusch S, Sahm F, Czabanka M, Luger S, et al. mTOR target NDRG1 confers MGMT-dependent resistance to alkylating chemotherapy. Proc Natl Acad Sci U S A. 2014 Jan 7; 111(1):409–14. [PubMed: 24367102]
76. Sommer EM, Dry H, Cross D, Guichard S, Davies BR, Alessi DR. Elevated SGK1 predicts resistance of breast cancer cells to Akt inhibitors. Biochem J. 2013 Jun 15; 452(3):499–508. [PubMed: 23581296]
77. Bakker WJ, Harris IS, Mak TW. FOXO3a is activated in response to hypoxic stress and inhibits HIF1-induced apoptosis via regulation of CITED2. Mol Cell. 2007 Dec 28; 28(6):941–53. [PubMed: 18158893]
78. Morrison MM, Young CD, Wang S, Sobolik T, Sanchez VM, Hicks DJ, et al. mTOR Directs Breast Morphogenesis through the PKC-alpha-Rac1 Signaling Axis. PLoS Genet Public Library of Science. 2015 Jul 1.11(7):e1005291.
79. Lee K, Nam KT, Cho SH, Gudapati P, Hwang Y, Park D-S, et al. Vital roles of mTOR complex 2 in Notch-driven thymocyte differentiation and leukemia. J Exp Med. 2012 Apr 9; 209(4):713–28. [PubMed: 22473959]
80. Cheng H, Zou Y, Ross JS, Wang K, Liu X, Halmos B, et al. RICTOR amplification defines a novel subset of lung cancer patients who may benefit from treatment with mTOR1/2 inhibitors. Cancer Discov. 2015 Sep 14.
81. Morrison-Joly M, Hicks DJ, Jones B, Sanchez V, Estrada MV, Young C, et al. Rictor/mTORC2 drives progression and therapeutic resistance of HER2-amplified breast cancers. Cancer Res. 2016 Apr.25:0008–5472. CAN – 15–3393 –.
82. Balko JM, Giltnane JM, Wang K, Schwarz LJ, Young CD, Cook RS, et al. Molecular profiling of the residual disease of triple-negative breast cancers after neoadjuvant chemotherapy identifies actionable therapeutic targets. Cancer Discov. 2014 Feb; 4(2):232–45. [PubMed: 24356096]

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

83. Masri J, Bernath A, Martin J, Jo OD, Vartanian R, Funk A, et al. mTORC2 activity is elevated in gliomas and promotes growth and cell motility via overexpression of rictor. Cancer Res. 2007 Dec 15; 67(24):11712–20. [PubMed: 18089801]
84. Sarbassov DD, Ali SM, Sengupta S, Sheen J-H, Hsu PP, Bagley AF, et al. Prolonged rapamycin treatment inhibits mTORC2 assembly and Akt/PKB. Mol Cell. 2006 Apr 21; 22(2):159–68. [PubMed: 16603397]
85. Benjamin D, Colombi M, Moroni C, Hall MN. Rapamycin passes the torch: a new generation of mTOR inhibitors. Nat Rev Drug Discov. 2011 Nov; 10(11):868–80. [PubMed: 22037041]
86. Hudes G, Carducci M, Tomczak P, Dutcher J, Figlin R, Kapoor A, et al. Temsirolimus, interferon alfa, or both for advanced renal-cell carcinoma. N Engl J Med. 2007 May 31; 356(22):2271–81. [PubMed: 17538086]
87. Yu K, Toral-Barza L, Shi C, Zhang W-G, Lucas J, Shor B, et al. Biochemical, cellular, and in vivo activity of novel ATP-competitive and selective inhibitors of the mammalian target of rapamycin. Cancer Res. 2009 Aug 1; 69(15):6232–40. [PubMed: 19584280]
88. Sun S-Y, Rosenberg LM, Wang X, Zhou Z, Yue P, Fu H, et al. Activation of Akt and eIF4E survival pathways by rapamycin-mediated mammalian target of rapamycin inhibition. Cancer Res. 2005 Aug 15; 65(16):7052–8. [PubMed: 16103051]
89. O'Reilly KE, Rojo F, She Q-B, Solit D, Mills GB, Smith D, et al. mTOR inhibition induces upstream receptor tyrosine kinase signaling and activates Akt. Cancer Res. 2006 Feb 1; 66(3): 1500–8. [PubMed: 16452206]
90. Hsieh AC, Costa M, Zollo O, Davis C, Feldman ME, Testa JR, et al. Genetic dissection of the oncogenic mTOR pathway reveals druggable addiction to translational control via 4EBP-eIF4E. Cancer Cell. 2010 Mar 16; 17(3):249–61. [PubMed: 20227039]
91. Naing A, Aghajanian C, Raymond E, Olmos D, Schwartz G, Oelmann E, et al. Safety, tolerability, pharmacokinetics and pharmacodynamics of AZD8055 in advanced solid tumours and lymphoma. Br J Cancer Cancer Research UK. 2012 Sep 25; 107(7):1093–9.
92. Basu B, Dean E, Puglisi M, Greystoke A, Ong M, Burke W, et al. First-in-Human Pharmacokinetic and Pharmacodynamic Study of the Dual m-TORC 1/2 Inhibitor AZD2014. Clin Cancer Res. 2015 Aug 1; 21(15):3412–9. [PubMed: 25805799]
93. Rodrik-Outmezguine VS, Okaniwa M, Yao Z, Novotny CJ, McWhirter C, Banaji A, et al. Overcoming mTOR resistance mutations with a new-generation mTOR inhibitor. Nature. 2016 Jun 9; 534(7606):272–6. [PubMed: 27279227]
94. Britten CD, Adjei AA, Millham R, Houk BE, Borzillo G, Pierce K, et al. Phase I study of PF-04691502, a small-molecule, oral, dual inhibitor of PI3K and mTOR, in patients with advanced cancer. Invest New Drugs. 2014 Jun; 32(3):510–7. [PubMed: 24395457]
95. Jänne PA, Cohen RB, Laird AD, Macé S, Engelman JA, Ruiz-Soto R, et al. Phase I safety and pharmacokinetic study of the PI3K/mTOR inhibitor SAR245409 (XL765) in combination with erlotinib in patients with advanced solid tumors. J Thorac Oncol. 2014 Mar; 9(3):316–23. [PubMed: 24496004]
96. Viñals F, Chambard JC, Pouysségur J. p70 S6 kinase-mediated protein synthesis is a critical step for vascular endothelial cell proliferation. J Biol Chem. 1999 Sep 17; 274(38):26776–82. [PubMed: 10480882]
97. Guba M, von Breitenbuch P, Steinbauer M, Koehl G, Flegel S, Hornung M, et al. Rapamycin inhibits primary and metastatic tumor growth by antiangiogenesis: involvement of vascular endothelial growth factor. Nat Med Nature Publishing Group. 2002 Feb 1; 8(2):128–35.
98. Hudson CC, Liu M, Chiang GG, Otterness DM, Loomis DC, Kaper F, et al. Regulation of Hypoxia-Inducible Factor 1 Expression and Function by the Mammalian Target of Rapamycin. Mol Cell Biol. 2002 Oct 15; 22(20):7004–14. [PubMed: 12242281]
99. Bernardi R, Guernah I, Jin D, Grisendi S, Alimonti A, Teruya-Feldstein J, et al. PML inhibits HIF-1alpha translation and neoangiogenesis through repression of mTOR. Nature. 2006 Aug 17; 442(7104):779–85. [PubMed: 16915281]
100. Sun S, Chen S, Liu F, Wu H, McHugh J, Bergin IL, et al. Constitutive Activation of mTORC1 in Endothelial Cells Leads to the Development and Progression of Lymphangiosarcoma through VEGF Autocrine Signaling. Cancer Cell. 2015 Dec 14; 28(6):758–72. [PubMed: 26777415]

Author Manuscript
Author Manuscript
Author Manuscript
Author Manuscript

101. Wang S, Amato KR, Song W, Youngblood V, Lee K, Boothby M, et al. Regulation of endothelial cell proliferation and vascular assembly through distinct mTORC2 signaling pathways. Mol Cell Biol. 2015 Apr; 35(7):1299–313. [PubMed: 25582201]
102. Guertin DA, Stevens DM, Thoreen CC, Burds AA, Kalaany NY, Moffat J, et al. Ablation in mice of the mTORC components raptor, rictor, or mLST8 reveals that mTORC2 is required for signaling to Akt-FOXO and PKCalpha, but not S6K1. Dev Cell. 2006 Dec; 11(6):859–71. [PubMed: 17141160]
103. Farhan MA, Carmine-Simmen K, Lewis JD, Moore RB, Murray AG. Endothelial Cell mTOR Complex-2 Regulates Sprouting Angiogenesis. PLoS One Public Library of Science. 2015 Jan 21.10(8):e0135245.
104. Guo F, Wang Y, Liu J, Mok SC, Xue F, Zhang W. CXCL12/CXCR4: a symbiotic bridge linking cancer cells and their stromal neighbors in oncogenic communication networks. Oncogene Nature Publishing Group. 2016 Feb 18; 35(7):816–26.
105. Phung TL, Ziv K, Dabydeen D, Eyiah-Mensah G, Riveros M, Perruzzi C, et al. Pathological angiogenesis is induced by sustained Akt signaling and inhibited by rapamycin. Cancer Cell. 2006 Aug; 10(2):159–70. [PubMed: 16904613]
106. Zhuang G, Yu K, Jiang Z, Chung A, Yao J, Ha C, et al. Phosphoproteomic analysis implicates the mTORC2-FoxO1 axis in VEGF signaling and feedback activation of receptor tyrosine kinases. Sci Signal. American Association for the Advancement of Science. 2013; 6(271):ra25.
107. Paik J-H, Kollipara R, Chu G, Ji H, Xiao Y, Ding Z, et al. FoxOs are lineage-restricted redundant tumor suppressors and regulate endothelial cell homeostasis. Cell. 2007 Jan 26; 128(2):309–23. [PubMed: 17254969]
108. Wilhelm, K., Happel, K., Eelen, G., Schoors, S., Oellerich, MF., Lim, R., et al. Nature. Nature Publishing Group, a division of Macmillan Publishers Limited. All Rights Reserved; 2016 Jan 6. FOXO1 couples metabolic activity and growth state in the vascular endothelium. advance on
109. Roy D, Sin S-H, Lucas A, Venkataramanan R, Wang L, Eason A, et al. mTOR inhibitors block Kaposi sarcoma growth by inhibiting essential autocrine growth factors and tumor angiogenesis. Cancer Res. 2013 Apr 1; 73(7):2235–46. [PubMed: 23382046]
110. Chatterjee S, Heukamp LC, Siobal M, Schöttle J, Wieczorek C, Peifer M, et al. Tumor VEGF:VEGFR2 autocrine feed-forward loop triggers angiogenesis in lung cancer. J Clin Invest American Society for Clinical Investigation. 2013 Apr 1; 123(4):1732–40.
111. Lane HA, Wood JM, McSheehy PMJ, Allegrini PR, Boulay A, Brueggen J, et al. mTOR inhibitor RAD001 (everolimus) has antiangiogenic/vascular properties distinct from a VEGFR tyrosine kinase inhibitor. Clin Cancer Res. 2009 Mar 1; 15(5):1612–22. [PubMed: 19223496]
112. Shinohara ET, Cao C, Niermann K, Mu Y, Zeng F, Hallahan DE, et al. Enhanced radiation damage of tumor vasculature by mTOR inhibitors. Oncogene. 2005 Aug 18; 24(35):5414–22. [PubMed: 15940265]
113. Fokas E, Im JH, Hill S, Yameen S, Stratford M, Beech J, et al. Dual inhibition of the PI3K/mTOR pathway increases tumor radiosensitivity by normalizing tumor vasculature. Cancer Res. 2012 Jan 1; 72(1):239–48. [PubMed: 22108822]
114. Sharma P, Allison JP, Curtin JA, Fridlyand J, Kageshita T, Patel HN, et al. The future of immune checkpoint therapy. Science American Association for the Advancement of Science. 2015 Apr 3; 348(6230):56–61.
115. Lastwika KJ, Wilson W, Li QK, Norris J, Xu H, Ghazarian SR, et al. Control of PD-L1 Expression by Oncogenic Activation of the AKT-mTOR Pathway in Non-Small Cell Lung Cancer. Cancer Res. 2016 Jan 15; 76(2):227–38. [PubMed: 26637667]
116. Kleffel S, Posch C, Barthel SR, Mueller H, Schlapbach C, Guenova E, et al. Melanoma Cell-Intrinsic PD-1 Receptor Functions Promote Tumor Growth. Cell. 2015 Sep 10; 162(6):1242–56. [PubMed: 26359984]
117. Granot Z, Fridlender ZG. Plasticity beyond cancer cells and the “immunosuppressive switch”. Cancer Res. 2015 Nov 1; 75(21):4441–5. [PubMed: 26475869]
118. Delgoffe GM, Kole TP, Zheng Y, Zarek PE, Matthews KL, Xiao B, et al. The mTOR kinase differentially regulates effector and regulatory T cell lineage commitment. Immunity. 2009 Jun 19; 30(6):832–44. [PubMed: 19538929]

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

119. Lee K, Gudapati P, Dragovic S, Spencer C, Joyce S, Killeen N, et al. Mammalian target of rapamycin protein complex 2 regulates differentiation of Th1 and Th2 cell subsets via distinct signaling pathways. Immunity. 2010 Jun 25; 32(6):743–53. [PubMed: 20620941]
120. Heikamp EB, Patel CH, Collins S, Waickman A, Oh M-H, Sun I-H, et al. The AGC kinase SGK1 regulates TH1 and TH2 differentiation downstream of the mTORC2 complex. Nat Immunol. 2014 May; 15(5):457–64. [PubMed: 24705297]
121. Pollizzi KN, Patel CH, Sun I-H, Oh M-H, Waickman AT, Wen J, et al. mTORC1 and mTORC2 selectively regulate $CD8^+$ T cell differentiation. J Clin Invest. 2015 May; 125(5):2090–108. [PubMed: 25893604]
122. Araki K, Turner AP, Shaffer VO, Gangappa S, Keller SA, Bachmann MF, et al. mTOR regulates memory CD8 T-cell differentiation. Nature. 2009 Jul 2; 460(7251):108–12. [PubMed: 19543266]
123. Zhang L, Tschumi BO, Lopez-Mejia IC, Oberle SG, Meyer M, Samson G, et al. Mammalian Target of Rapamycin Complex 2 Controls CD8 T Cell Memory Differentiation in a Foxo1-Dependent Manner. Cell Rep. 2016; 14(5):1206–17. [PubMed: 26804903]
124. Byles V, Covarrubias AJ, Ben-Sahra I, Lamming DW, Sabatini DM, Manning BD, et al. The TSC-mTOR pathway regulates macrophage polarization. Nat Commun. 2013; 4:2834. [PubMed: 24280772]
125. Lee K, Heffington L, Jellusova J, Nam KT, Raybuck A, Cho SH, et al. Requirement for Rictor in homeostasis and function of mature B lymphoid cells. Blood American Society of Hematology. 2013 Oct 3; 122(14):2369–79.
126. LaGory EL, Giaccia AJ. The ever-expanding role of HIF in tumour and stromal biology. Nat Cell Biol. Nature Publishing Group, a division of Macmillan Publishers Limited. All Rights Reserved. 2016 Mar 30; 18(4):356–65.
127. Mayerhofer M, Valent P, Sperr WR, Griffin JD, Sillaber C. BCR/ABL induces expression of vascular endothelial growth factor and its transcriptional activator, hypoxia inducible factor-1alpha, through a pathway involving phosphoinositide 3-kinase and the mammalian target of rapamycin. Blood American Society of Hematology. 2002 Nov 15; 100(10):3767–75.
128. Brugarolas JB, Vazquez F, Reddy A, Sellers WR, Kaelin WG. TSC2 regulates VEGF through mTOR-dependent and -independent pathways. Cancer Cell. 2003 Aug; 4(2):147–58. [PubMed: 12957289]
129. Mateo J, Olmos D, Dumez H, Poondru S, Samberg NL, Barr S, et al. A first in man, dose-finding study of the mTORC1/mTORC2 inhibitor OSI-027 in patients with advanced solid malignancies. Br J Cancer. 2016 Apr 12; 114(8):889–96. [PubMed: 27002938]
130. Powles T, Wheater M, Din O, Geldart T, Boleti E, Stockdale A, et al. A Randomised Phase 2 Study of AZD2014 Versus Everolimus in Patients with VEGF-Refractory Metastatic Clear Cell Renal Cancer. Eur Urol. 2016 Mar; 69(3):450–6. [PubMed: 26364551]
131. Bendell JC, Kelley RK, Shih KC, Grabowsky JA, Bergsland E, Jones S, et al. A phase I dose-escalation study to assess safety, tolerability, pharmacokinetics, and preliminary efficacy of the dual mTORC1/mTORC2 kinase inhibitor CC-223 in patients with advanced.
132. Bendell JC, Kurkjian C, Infante JR, Bauer TM, Burris HA, Greco FA, et al. A phase 1 study of the sachet formulation of the oral dual PI3K/mTOR inhibitor BEZ235 given twice daily (BID) in patients with advanced solid tumors. Invest New Drugs. 2015 Apr; 33(2):463–71. [PubMed: 25707361]
133. Seront E, Rottey S, Filleul B, Glorieux P, Goeminne J, Verschaeve V, et al. Phase II study of dual phosphoinositol-3-kinase (PI3K) and mammalian target of rapamycin (mTOR) inhibitor BEZ235 in patients with locally advanced or metastatic transitional cell carcinoma (TCC). BJU Int. 2016 Jan 18.
134. Fazio N, Buzzoni R, Baudin E, Antonuzzo L, Hubner RA, Lahner H, et al. A Phase II Study of BEZ235 in Patients with Everolimus-resistant, Advanced Pancreatic Neuroendocrine Tumours. Anticancer Res. 2016 Feb; 36(2):713–9. [PubMed: 26851029]
135. Wen PY, Omuro A, Ahluwalia MS, Fathallah-Shaykh HM, Mohile N, Lager JJ, et al. Phase I dose-escalation study of the PI3K/mTOR inhibitor voxtalisib (SAR245409, XL765) plus temozolomide with or without radiotherapy in patients with high-grade glioma. Neuro Oncol. 2015 Sep; 17(9):1275–83. [PubMed: 26019185]

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

136. Papadopoulos KP, Egile C, Ruiz-Soto R, Jiang J, Shi W, Bentzien F, et al. Efficacy, safety, pharmacokinetics and pharmacodynamics of SAR245409 (voxtalisib, XL765), an orally administered phosphoinositide 3-kinase/mammalian target of rapamycin inhibitor: a phase 1 expansion cohort in patients with relapsed or refractory lymphoma. Leuk Lymphoma. 2015 Jun; 56(6):1763–70. [PubMed: 25300944]
137. Papadopoulos KP, Tabernero J, Markman B, Patnaik A, Tolcher AW, Baselga J, et al. Phase I safety, pharmacokinetic, and pharmacodynamic study of SAR245409 (XL765), a novel, orally administered PI3K/mTOR inhibitor in patients with advanced solid tumors. Clin Cancer Res. 2014 May 1; 20(9):2445–56. [PubMed: 24583798]
138. Dolly SO, Wagner AJ, Bendell JC, Kindler HL, Krug LM, Seiwert TY, et al. Phase I Study of Apitolisib (GDC-0980), Dual Phosphatidylinositol-3-Kinase and Mammalian Target of Rapamycin Kinase Inhibitor, in Patients with Advanced Solid Tumors. Clin Cancer Res. 2016 Jan 19.
139. Powles T, Lackner MR, Oudard S, Escudier B, Ralph C, Brown JE, et al. Randomized Open-Label Phase II Trial of Apitolisib (GDC-0980), a Novel Inhibitor of the PI3K/Mammalian Target of Rapamycin Pathway, Versus Everolimus in Patients With Metastatic Renal Cell Carcinoma. J Clin Oncol. 2016 May 10; 34(14):1660–8. [PubMed: 26951309]
140. Shapiro GI, Bell-McGuinn KM, Molina JR, Bendell J, Spicer J, Kwak EL, et al. First-inHuman Study of PF-05212384 (PKI-587), a Small-Molecule, Intravenous, Dual Inhibitor of PI3K and mTOR in Patients with Advanced Cancer. Clin Cancer Res. 2015 Apr 15; 21(8):1888–95. [PubMed: 25652454]
141. Munster P, Aggarwal R, Hong D, Schellens JHM, van der Noll R, Specht J, et al. First-inHuman Phase I Study of GSK2126458, an Oral Pan-Class I Phosphatidylinositol-3-Kinase Inhibitor, in Patients with Advanced Solid Tumor Malignancies. Clin Cancer Res. 2015 Nov 24; 22(8):1932–9. [PubMed: 26603258]
142. Del Campo JM, Birrer M, Davis C, Fujiwara K, Gollerkeri A, Gore M, et al. A randomized phase II non-comparative study of PF-04691502 and gedatolisib (PF-05212384) in patients with recurrent endometrial cancer. Gynecol Oncol. 2016 Apr 18.
143. Mahadevan D, Chiorean EG, Harris WB, Von Hoff DD, Stejskal-Barnett A, Qi W, et al. Phase I pharmacokinetic and pharmacodynamic study of the pan-PI3K/mTORC vascular targeted pro-drug SF1126 in patients with advanced solid tumours and B-cell malignancies. Eur J Cancer. 2012 Dec; 48(18):3319–27. [PubMed: 22921184]
144. Markman B, Tabernero J, Krop I, Shapiro GI, Siu L, Chen LC, et al. Phase I safety, pharmacokinetic, and pharmacodynamic study of the oral phosphatidylinositol-3-kinase and mTOR inhibitor BGT226 in patients with advanced solid tumors. Ann Oncol. 2012 Sep; 23(9): 2399–408. [PubMed: 22357447]

Author Manuscript
Author Manuscript
Author Manuscript
Author Manuscript

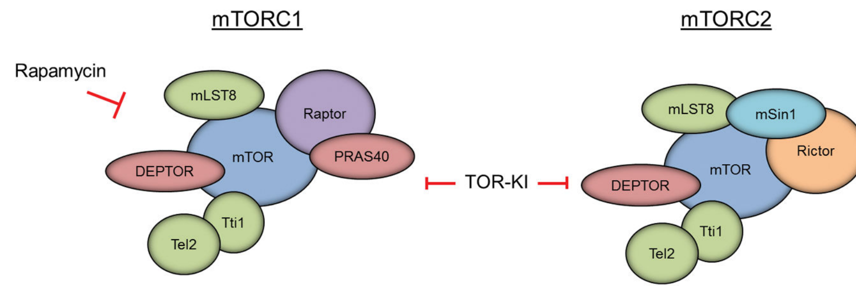

Figure 1. Schematic representation of mTOR complexes

mTORC1 consists of the mTOR kinase, mLST8, DEPTOR, Tti/Tel2, Raptor, and PRAS40. mTORC2 also shares the mTOR kinase, mLST8, Tti/Tel2, and DEPTOR, but contains unique components Rictor and mSin1. Rapamycin is a known allosteric inhibitor of mTORC1, while TOR kinase inhibitors (TOR-KIs) inhibit the activities of both complexes.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

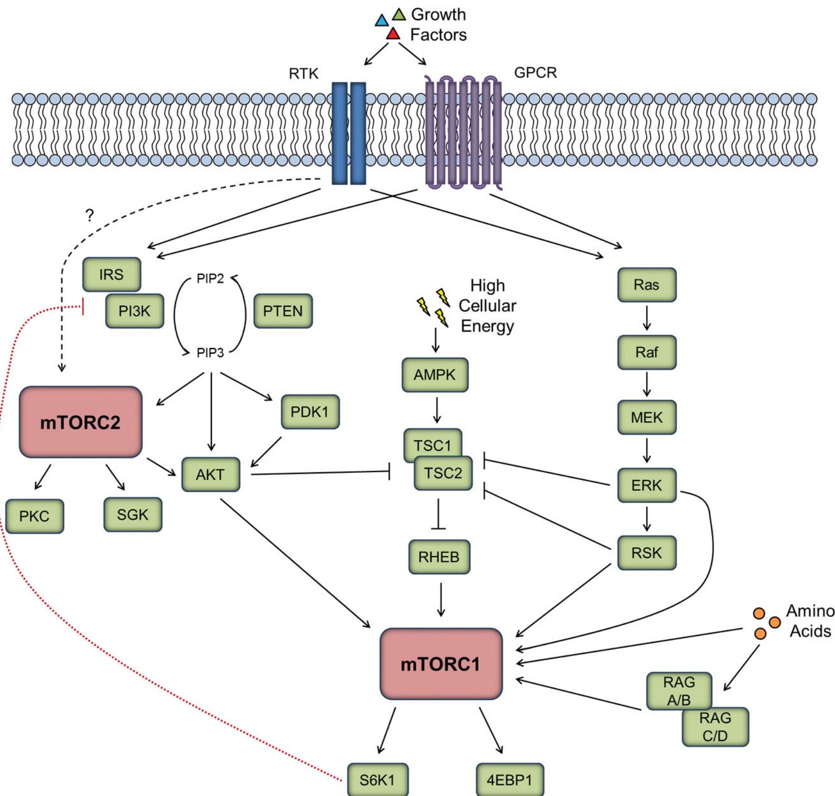

Figure 2. Overview of the mTOR signaling pathway

mTOR signaling is activated by a variety of environmental cues including growth factors, high cellular energy, and amino acids. Growth factors activate both mTORC1 and mTORC2, through binding of receptor tyrosine kinases (RTKs) or G-protein coupled receptors (GPCRs) and activation of PI3K or Ras-MAPK signaling cascades. PI3K phosphorylates PIP2 to increase the amount of PIP3 in the membrane, allowing co-localization of AKT, PDK1 and mTORC2. PDK1 phosphorylates AKT at T308, while mTORC2 phosphorylates AKT at S473 for complete activation. AKT in turn activates mTORC1 by inhibiting TSC2, a GAP for RHEB, an activator of mTORC1. AKT phosphorylation of PRAS40 promotes its dissociation from mTORC1 for full activation. ERK and RSK, both part of the Ras-MAPK signaling pathway, can also inhibit TSC2 to activate mTORC1 or activate mTORC1 directly through phosphorylation of PRAS40. High ATP levels in the cell inhibit AMPK, an activator of TSC2, thereby increasing the activities of RHEB and mTORC1. Intra-lysosomal arginine and cytoplasmic leucine stimulate Rag-dependent localization of mTORC1 to the lysosome where RHEB can activate mTORC1. Cytoplasmic glutamine triggers lysosomal localization of mTORC1 through a Rag-independent mechanism. Downstream targets of mTORC1 include S6K1 and 4EBP1, while downstream targets of mTORC2 include AKT, PKC, and SGK. S6K1 inhibits PI3K, completing a negative feedback loop on AKT signaling.

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Table 1

mTOR inhibitors approved by the FDA for cancer treatment

Drug	Date of approval	Cancer Type	Therapeutic Condition	Marketed By
Rapamycin/Sirolimus (Rapamune)	5.29.2015	Lymphangioleiomyomatosis	Monotherapy	Pfizer (Wyeth)
Temsirolimus (Torisel)	5.30.2007	Renal Cell Carcinoma	Monotherapy	Pfizer (Wyeth)
Everolimus (Afinitor)	5.30.2009	Advanced Renal Cell Carcinoma	Monotherapy	Novartis
	10.29.2010	Subependymal Giant Cell Astrocytoma (SEGA) associated with Tuberous Sclerosis Complex (TSC)	Monotherapy	
	5.5.2011	Progressive Neuroendocrine Tumors of Pancreatic Origin	Monotherapy	
	7.20.2012	Hormone Receptor Positive, HER2 Negative Breast Cancer	In combination with Exemestane	
	8.29.2012	Pediatric and Adult SEGA associated with TSC	Monotherapy	
	2.26.2016	Neuroendocrine Tumors of Gastrointestinal or Lung Origin	Monotherapy	

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Table 2

mTOR inhibitors currently in clinical trials

Drug Target	Drug Name	Published Clinical Trial	Cancer Type	References	Active Clinical Trials*	Combination Therapy
mTOR kinase inhibitor	OSI-027	Phase I	Advanced solid tumors	129		
	AZD2014	Phase I/II	Advanced solid tumors, clear cell renal cancer	92,130	18	• Paclitaxel • AZD5363 • Selumetinib • Palbociclib • fulvestrant • rituximab • anastrozole • olaparib
	AZD8055	Phase I	Advanced solid tumors, lymphoma	91		
	CC223	Phase I	Advanced solid tumors, multiple myeloma	131	2	• CC122 or CC292 +/− Rituxumab
	MLN0128	None available			10	• Paclitaxel • Bevacizumab • MLN1117 • Alisertib or Paclitaxel or Cetuximab or Irenotecan • Exemestane or Fulvestrant
PI3K/mTOR dual inhibitor	BEZ235	Phase I/II	Advanced solid tumors, transitional cell carcinoma, pancreatic neuroendocrine tumors	132–134	1	
	XL765/ SAR254409	Phase I	Advanced solid tumors, high-grade glioma, lymphoma	95,135–137		
	GDC0980	Phase I/II	Advanced solid tumors, metastatic renal cell carcinoma	138,139	2	• Fulvestrant • Abiraterone Acetate
	PKI587	Phase I	Advanced solid	140	3	• Carboplatin and paclitaxel

Drug Target	Drug Name	Published Clinical Trial	Cancer Type	References	Active Clinical Trials*	Combination Therapy
						• Docetaxel or Cisplatin or Dacomitinib • Palbociclib and Faslodex
	GSK2126458	Phase I	Advanced solid tumors	141		
	PF04691502	Phase I/II	Advanced solid tumors, endometrial cancer	94,142		
	SF1126	Phase I	Advanced solid tumors, B-cell malignancies	143	1	
	BGT226	Phase I	Advanced solid tumors	144		

*Data summarized from ClinicalTrials.gov

Author Manuscript Author Manuscript Author Manuscript Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Table 3

Function of mTORC1 and mTORC2 in tumor and endothelial cells

Tumor Cell		Endothelial Cell	
mTORC1			
Cell Size and Proliferation	Refs. 36,37	Autocrine VEGF signaling through HIF-1 α translation	Refs. 100,109,110
Metabolic Reprogramming: glycolysis, glutaminolysis, nucleotide synthesis, lipogenesis	Refs. 50–57, 59	Vessel Permeability	Ref. 101
Stimulation of Angiogenesis	Refs. 97–99,	Cell Proliferation	Ref. 96,100
mTORC2			
Cell Survival	Ref.71	Vessel Morphology and Permeability	Refs. 101,105
Metabolic reprogramming: glycolysis, hypoxic response	Refs. 73,77	Cell Proliferation and Vascular Assembly	Ref. 101
Chemotherapy Resistance	Ref. 75	Metabolic Activity	Ref.106–108