

Roles of Rictor alterations in gastrointestinal tumors (Review)

RUIZHEN CAO^{1,2*}, SHUILONG GUO^{1*}, LI MIN¹ and PENG LI¹

¹Department of Gastroenterology, Beijing Friendship Hospital, Capital Medical University, National Clinical Research Center for Digestive Disease, Beijing Digestive Disease Center, Beijing Key Laboratory for Precancerous Lesion of Digestive Disease, Beijing 100050; ²Department of Gastroenterology, Ordos Central Hospital, Ordos School of Clinical Medicine, Inner Mongolia Medical University, Ordos, Inner Mongolia 017000, P.R. China

Received April 5, 2023; Accepted November 28, 2023

DOI: 10.3892/or.2024.8696

Abstract. Gastrointestinal tumors account for five of the top 10 causes of mortality from all cancers (colorectal, liver, stomach, esophageal and pancreatic cancer). Mammalian target of rapamycin (mTOR) signaling is commonly dysregulated in various human cancers. As a core component of the mTOR complex 2 (mTORC2), Rictor is a key effector molecule of the PI3K/Akt pathway. A high alteration rate of Rictor has been observed in gastrointestinal tumors, and such Rictor alterations are often associated with resistance to chemotherapy and related adverse clinical outcomes. However, the exact roles of Rictor in gastrointestinal tumors remain elusive. The aim of the present study was to critically discuss the following: i) Mutation and biological characteristics of Rictor in tumors with a detailed overview of Rictor in cell proliferation, angiogenesis, apoptosis, autophagy and drug resistance; ii) the role of Rictor in tumors of the digestive system, particularly colorectal, hepatobiliary, gastric, esophageal and pancreatic cancer and cholangiocarcinoma; and iii) the current status and prospects of targeted therapy for Rictor by inhibiting Akt activation. Despite the growing realization of the importance of Rictor/mTORC2 in cancer, the underlying mechanistic details remain poorly understood; this needs to change in order for the development of efficient targeted therapies and re-sensitization of therapy-resistant cancers to be made possible.

Contents

1. Introduction
2. Overview of Rictor and the mTOR pathway
3. General biological effects of Rictor in cancer cells
4. Effects of Rictor in gastrointestinal cancers
5. Targeted therapy
6. Conclusion and future perspectives

1. Introduction

According to the most recent figures from the International Agency for Research on Cancer on incidence and mortality, there will be 28.4 million new cases of cancer worldwide in 2040, a 47% increase from 2020. Out of all cancer cases, gastrointestinal tumors account for five of the top 10 causes of mortality [colorectal cancer (CRC; 9.4%), liver cancer (8.3%), stomach cancer (7.7%), esophageal cancer (5.5%), and pancreatic cancer (4.7%)] (1). Owing to the high metastasis and recurrence of these gastrointestinal tumors, the 5-year overall survival rate for advanced tumors is poor (2,3). Cancer treatment has undergone profound changes in recent years with the continuous development of the understanding of cancer biology at the molecular level. For instance, a large number of targeted drugs have been approved as a first-line treatment for numerous tumors (4). However, clinical studies revealed that these drugs are ineffective for patients with Rictor alterations (5). These studies suggested that Rictor is involved in tumor resistance and may act as a therapeutic target.

Genomic instability and mutation are important features of cancer cells. According to The Cancer Genome Atlas (TCGA) database (6), as determined by the alterations of Rictor in respective patient samples as a fraction of the total number of patients screened, in non-small-cell lung cancer, 41.3% (19/46 cases) of patients had altered Rictor levels. Similarly, the figures for altered Rictor are 14.11% (23/163 cases) in esophagogastric cancer, 13.64% (42/308 cases) in pancreatic cancer, 11.54% (23/163 cases) in CRC and 8% (29/358 cases) in hepatobiliary cancer (Fig. 1). Further data from the TCGA dataset comprising 991 samples showed different types of Rictor alterations (Fig. 2). In addition, the overall survival rates of patients with high Rictor expression in tumor tissues was observed to be low. For instance, Bian *et al* (7) demonstrated

Correspondence to: Professor Peng Li, Department of Gastroenterology, Beijing Friendship Hospital, Capital Medical University, National Clinical Research Center for Digestive Disease, Beijing Digestive Disease Center, Beijing Key Laboratory for Precancerous Lesion of Digestive Disease, 95 Yong'an Road, Xicheng District, Beijing 100050, P.R. China
E-mail: lipeng@ccmu.edu.cn

*Contributed equally

Key words: gastrointestinal tumors, mTOR, Rictor, PI3K, Akt

through immunohistochemistry that high Rictor expression is associated with rapid tumor progression and poor prognosis in patients with gastric cancer (GC). A study from the Southern Medical University of China revealed that a high Rictor expression leads to poor clinical prognosis in CRC (8). From the analysis of 201 samples of esophageal squamous cell carcinoma (ESCC), it was found that Rictor expression was positively associated ($P=0.011$) with the cancer stage, according to the grading by the American Joint Committee on Cancer (AJCC), and negatively associated ($P=0.007$) with survival (9). These findings are sufficiently exciting to warrant a detailed discussion of the role of Rictor in the biology of gastrointestinal cancers.

2. Overview of Rictor and the mTOR pathway

The mTOR and its signaling pathway have important roles in regulating protein synthesis, cell growth, apoptosis, angiogenesis and migration. Dysfunction of the mTOR signaling pathway is common in several human cancers (10,11). mTOR exists in two complexes: The mTOR complex 1 (mTORC1) and mTORC2. mTORC2 consists of mTOR, mTOR-associated protein, LST8 homolog mLST8, Rictor, mSin1 and proteins associated with Rictor 1/2, which are sensitive to growth factor levels and responsible for the regulation of cell proliferation, metabolism, survival and cytoskeletal remodeling (12). Rictor is a core subunit of mTORC2. The function of mTORC2 is dependent on Rictor, which is insensitive to rapamycin.

Rictor was discovered and characterized by Sarbassov *et al* (13). It has 1,709 amino acids with a molecular weight of 190 kDa. Rictor has seven domains with sequence conservation in mammals. It signals to the actin cytoskeleton by regulating protein kinase C α (PKC α) phosphorylation. A structural analysis and functional domain studies revealed that Rictor contains the HEAT and WD40 domains, which may be the common motifs interacting with mTORC (14). Rictor also has a pleckstrin homology domain that is similar to human 39S protein L17 and 50S protein L17. This ribosome-binding domain is required for cellular localization and transmission of signals to downstream targets by Rictor/mTORC2 interaction.

3. General biological effects of Rictor in cancer cells

Mutations and biological characteristics of Rictor in tumors reported to date are presented in Fig. 3.

Autophagy. Eukaryotes have used autophagy as a crucial intracellular turnover process throughout evolution. It enables cells to keep their intracellular environment stable. However, the influence of autophagy on specific cell functions remains controversial. Autophagy has been linked to cell survival and death processes under metabolic stress. Autophagy reportedly affects tumorigenesis and treatment (15,16). Using bioinformatics analyses, Hao *et al* (17) observed that Rictor was a direct target of microRNA (miR)-let-7a. Rescue experiments *in vitro* showed that miR-let-7a promoted the autophagy level by inhibiting the expression of Rictor in GC cells. In addition, as an upstream executor of the Akt-mTOR signaling pathway, Rictor exerted its effect on autophagy by phosphorylating Akt and mTOR, and this regulatory process was also mediated by

miR-let-7a. miR-let-7a in GC regulates autophagy by targeting Rictor and follows the regulation of the Akt-mTOR signaling pathway. Seo *et al* (18) reported that downregulation of Rictor was induced after co-treatment with PP242 and curcumin in renal cancer cells. Downregulation of Rictor increased cytosolic calcium release from the endoplasmic reticulum, leading to lysosomal damage in the cell, which induced autophagy. Liu *et al* (19) reported that Akt is further activated by triggering the phosphorylation of mTOR, which regulates the growth, autophagy and apoptosis of tumor cells, including GC cells.

Proliferation. The PI3K/Akt signaling pathway stimulates cell survival and metabolism, inhibits apoptosis and regulates tumor cell survival and proliferation. The activation of Akt depends on the phosphorylation of PIP3 (PDK1) at Thr308 and PDK2 at Ser473, and the phosphorylation of Ser473 promotes that of Thr308. Sarbassov *et al* (20) found that mTORC2 is PDK2 at the Ser473 site of phosphorylated Akt in *Drosophila* cells. Hresko and Mueckler (21) verified the above hypothesis in 3T3-L1 cells. These studies suggested that Rictor participates in the PI3K/Akt signaling pathway with mTORC2 and then regulates cell survival and nutrient uptake through mTORC1 downstream of Akt, as well as protein synthesis and cell cycle through glycogen synthase kinase 3 (GSK-3). The PI3K/Akt/mTOR signaling pathway is frequently altered in malignant tumors and Rictor is a key component of this pathway (22). Resistance to the inhibition of the adjacent PI3K pathway is usually characterized by the feedback activation of Akt, which is related to the mechanisms involving Rictor (23).

Serum and glucocorticoid-induced protein kinase (SGK) is a member of the protein kinase A/protein kinase G/protein kinase C (AGC) family and exists in three subtypes in cells: SGK1, SGK2 and SGK3. SGK1 is usually activated by insulin or nutritional factors and helps regulate cell nutrient uptake (24), survival, proliferation and apoptosis (25). García-Martínez *et al* (26) found that Rictor can directly bind to SGK1 in the form of mTORC2 and phosphorylate its Ser422 site, independent of PI3K. This finding has been verified in 293, MCF-7 and HeLa cells.

Apoptosis inhibition. Studies have confirmed that Rictor stimulates cell growth and proliferation by activating Akt (also known as protein kinase B), increasing the cells' resistance to apoptosis and promoting angiogenesis (27,28). Rictor overexpression in GC is associated with poor prognosis. In particular, Rictor activates caveolin 1 (Cav1) through the Akt signaling pathway to inhibit the apoptosis of GC cells (29). Liu *et al* (30) reported that real-time PCR and western blot showed that miR-153 downregulated the expression of Rictor, and this was related to the anti-tumor effect through increasing apoptosis and inhibiting the growth of breast cancer cells. A recent study (27) suggested that Rictor is a substrate for caspase-3 and is cleaved during apoptosis. In kidney cancer cells, Rictor silencing increases apoptosis and concomitantly enhances rasfonin-induced autophagy (31). In ESCC, the downregulation of Rictor expression inhibits proliferation and migration and induces ECa-109 and EC9706 cell cycle arrest and apoptosis (32). *In vitro* experiments

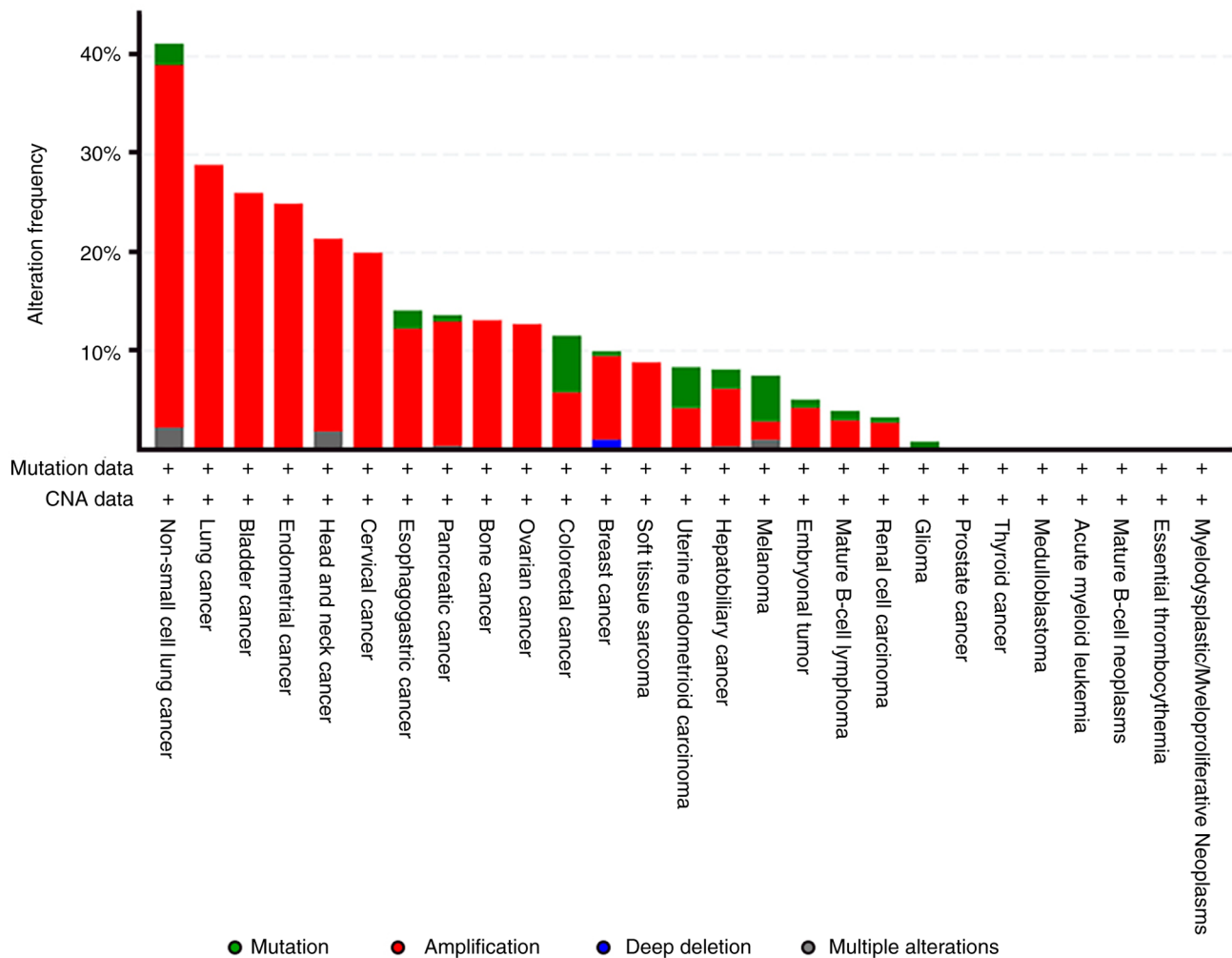


Figure 1. Alteration frequency of Rictor in different cancers (data from TCGA). Mutation data on Rictor is provided for different cancer types, as indicated. TCGA, The Cancer Genome Atlas; CNA, copy number alteration.

showed that Rictor knockdown suppressed the proliferation, inhibited the migration and invasion, and induced apoptosis of GC cells (33).

Angiogenesis. Rictor regulates the migration and proliferation of vascular endothelial cells, two events that are crucial for tumor angiogenesis. Wang *et al* (34) reported that Rictor deletion drastically reduced the vascular endothelial growth factor (VEGF)-induced proliferation and tubulogenesis of endothelial cells *in vitro* by inhibiting Akt activity through PKC α phosphorylation. Rictor/mTORC2 inhibits the prostaglandin E2-induced proliferation and migration of vascular endothelial cells by regulating Rac and Akt activation. The hypoxia-induced proliferation of endothelial cells depends on the involvement of Rictor/mTORC2 in regulating the angiogenic mimicry of melanoma through the Akt-MMP-2/9 pathway (35).

Rictor regulates VEGF expression in addition to controlling endothelial cell proliferation and migration (28). Guan *et al* (36) reported that the tumor suppressor miR-218 specifically targets Rictor to inhibit angiogenesis in prostate cancer, and this mechanism may be active in other cancer tissues, including gastrointestinal cancers. mTORC2 is a

key signaling point that promotes VEGF-mediated angiogenesis in vascular endothelial cells by regulating Akt and PKC α (37).

Cellular motility. The actin cytoskeleton and microtubules are the primary cellular structures that maintain cellular morphology and stress (38). Rictor regulates actin cytoskeleton remodeling through PKC, and PKC α is a representative of typical PKC. Sarbassov *et al* (20) found that the Rictor/mTOR complex can directly bind and phosphorylate PKC α to regulate the actin cytoskeleton and, consequently, cell motility. Guertin *et al* (39) demonstrated that Rictor binds to PKC α and regulates its phosphorylation in Raptor-, Rictor-, mLST8- and mTOR-knockout mice. PKC ζ , a representative of atypical PKC and has an important role in regulating actin cytoskeletal remodeling. Rictor can directly bind to PKC ζ near the cell membrane without mTORC2 in human breast cancer. In addition, the phosphorylation of PKC ζ and its downstream F-actin binding protein cofilin regulate actin remodeling and cell motility (40).

Rho GTPases with a molecular weight of 21 kDa are a family of small G proteins, including cell division control protein 42 homolog (Cdc42), Rac family small GTPase 1

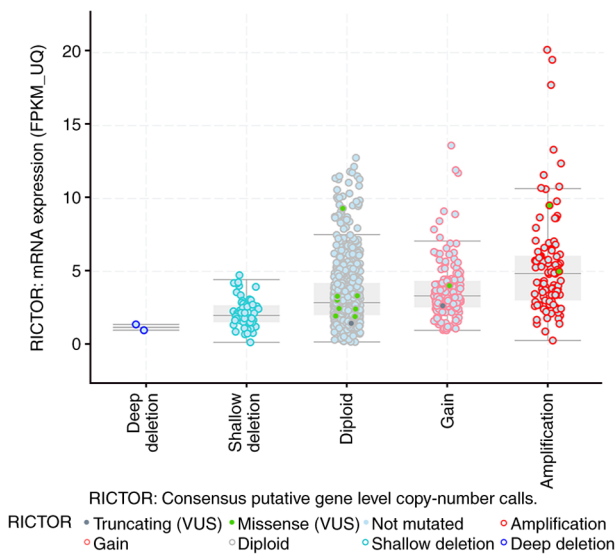


Figure 2. Consensus putative gene level copy-number calls of Rictor from 991 samples with data in both profiles (axes). Data are shown for Rictor mRNA expression vs. Rictor consensus putative gene level copy numbers. VUS, variant of uncertain significance.

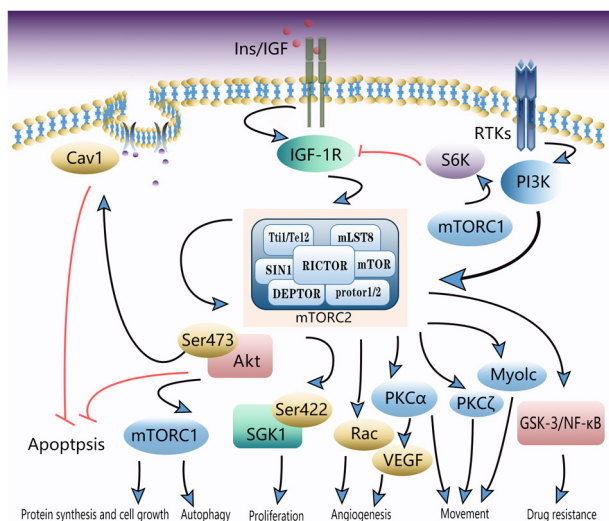


Figure 3. Schematic of Rictor signaling pathways that regulate tumor growth, survival, metastasis and drug resistance. As indicated, Rictor, as part of the mTORC2 pathway, has a central role in several signaling cascades that originate at the cell membrane level through various receptors. Once Rictor is involved, the effects are apparent with regulation of several downstream signaling molecules, with prominent ones being Akt, PKCs, SGK1, VEGF and NF- κ B. An overall enhancing effect on protein synthesis, cell growth, autophagy, cellular proliferation, angiogenesis, cell motility and drug resistance was observed. mTORC2, mammalian target of rapamycin complex 2; PKC, protein kinase C; SGK1, serum and glucocorticoid-induced protein kinase 1; VEGF, vascular endothelial growth factor.

(Rac1) and Ras homolog (Rho) family member A. These proteins are responsible for regulating actin remodeling, microtubule treadmilling and cell migration. Rictor can maintain or enhance Rac1/Cdc42 activity by regulating Rho GDP-dissociation inhibitor 2 (Rho GDI2), a suppressor of Rho-GDP. Thus, Rictor regulates actin remodeling and tumor cell motility by regulating Rho GDI2 through an mTOR-independent pathway (41).

Rictor also regulates actin remodeling through molecular motors, such as Myosin-1C (Myo1c). Agarwal *et al* (42) found that in 3T3-L1 fibroblastic cells, Rictor can directly bind to Myo1c, form a stable complex independent of mTORC2 and then regulate actin reconstruction by controlling the phosphorylation of paxillin Tyr18. This regulation and that of cell motility are not affected by mTORC2 or PI3K inhibitors.

Drug resistance. In patients with GC, higher expression of Rictor has been linked to tumor growth and poor prognosis (12). mTORC1 is sensitive to rapamycin treatment and mediates eukaryotic initiation factor 4E-binding protein-1 (4E-BP1), as well as the phosphorylation and activation of p70S6 ribosomal kinase (S6K) (43). In this light, treatment with rapamycin or its analogs was observed to inhibit the mTORC1/S6K pathway and reduce the negative feedback loop of insulin-like growth factor-1 receptor (IGF-1R) from S6K to IGF-1, impairing mTORC2 signaling through the complete pathway and leading to Akt activation (44). Furthermore, the paradoxical activation of Akt is undesirable, as it elicits drug resistance and cell survival, both of which are harmful to the effectiveness of mTORC1 inhibitor therapy. In other words, the mTOR inhibitor rapamycin could inhibit mTORC1 in cancer cells and may lead to Akt phosphorylation through mTORC2 activation. Lang *et al* (45) found that rapamycin upregulates IGF-IR and human epidermal growth factor receptor 2 (HER2) in GC and pancreatic cancer cells. Furthermore, mTORC2 has been shown to promote the activation of IGF-IR and insulin receptors by activating mTOR tyrosine kinase and participate in tumorigenesis (46). Rictor downregulation by RNA interference (RNAi) and the induction of receptor kinase expression are mediated by Akt activation induced by mTORC2. In addition, mTORC2 inhibition reduces the motility of cancer cells by suppressing GSK-3/NF- κ B activity (45).

4. Effects of Rictor in gastrointestinal cancers

CRC. More than 1.9 million new cases of CRC, including anal cancer, and 935,000 deaths are expected in the coming years (1). In general, CRC ranks third in incidence and second in mortality; the higher mortality is likely due to the development of drug resistance (47). Bellier *et al* (48) used methylglyoxal (MGO), a metabolite of glycolysis that promotes tumor growth and metastasis, to induce Akt activation and analyzed CRC resistance. The study found that MGO induces Akt activation by regulating PI3K/mTORC2 and heat shock protein (Hsp)27. The premise of that study was that cancers with Kirsten rat sarcoma viral oncogene homologue (KRAS) mutations exhibit poor response rates to therapies and that cells with mutated KRAS under MGO stress rely on Akt for their survival, particularly when compared to the cells with wild-type KRAS. Akt is activated through PI3K/mTORC2 and Hsp27. An important finding was that MGO scavengers can inhibit Akt, which may result in the re-sensitization of KRAS-mutant cells to cetuximab. In another study, the autophagy-related genes Beclin 1, Raptor and Rictor were shown to be associated with the development and progression of CRC and multidrug resistance (MDR) (49). All three genes were selected due

to their association with autophagy. Immunohistochemistry and reverse transcription-quantitative PCR-based evaluation was performed in 279 patients with CRC. These three autophagy-related genes, as well as light chain 3 (LC3) and MDR-1, were significantly upregulated in CRC tissues as compared with the adjacent control tissues. Furthermore, their expression in patients with lymph node metastasis was higher than that in patients without. LC3 was found to be positively correlated with Beclin 1 and Raptor and negatively correlated with Raptor and mTORC in patients with CRC. Furthermore, it was revealed that the five-year survival rate of patients with CRC without lymph node metastasis, positive/high expression of Rictor, Beclin 1 and LC3, and negative Raptor and mTOR expression, was higher than that of patients with lymph node metastasis, high Rictor, Beclin 1 and LC3 expression, and high Raptor and mTOR expression.

Hepatobiliary cancers. Hepatocellular carcinoma (HCC) is a leading cause of cancer-related mortality worldwide, due to the lack of precise therapeutic targets (50). mTORC2/Rictor is involved in the pretumor state of HCC and participates in the malignant transformation of liver HCC. Reyes-Gordillo *et al* (51) showed that Akt isomers are activated in alcoholic liver disease by increasing the expression levels of mTORC2 and genes associated with inflammation, proliferation and fibrosis. In addition, mTORC2 affects HCC tumorigenesis by regulating metabolic reprogramming. mTORC2 triggers the synthesis of fatty acids and lipids, resulting in liver steatosis and tumorigenesis (52). Xin *et al* (53) identified that Rictor interacted with *Homo sapiens* actin binding LIM protein 1 (ABLIM1) and regulated its serine phosphorylation in HCC cells. ABLIM1, as a previously unknown phosphorylation target of Rictor, induced by Rictor, was indicated to have an important role in controlling actin polymerization in HCC cells. Rictor knockdown significantly suppressed cell migration and actin polymerization. Immunohistochemical results showed that mTORC2 is activated in 60% of HCC cases (54). High mTORC2 expression was found to be associated with poor prognosis of patients with HCC.

Rictor knockdown was observed to inhibit the growth of HCC cells *in vitro* (55). In the liver, Akt has two subtypes: Akt1 and Akt2. In c-Myc-induced HCC, Akt1 is phosphorylated and activated by mTORC2. Akt1 is the main driver of HCC formation and its inhibition can prevent c-Myc tumor development (56). mTORC2 is also involved in liver cancer metastasis and drug resistance. Choline kinase α (CHKA) is an enzyme associated with liver cancer metastasis and epidermal growth factor receptor resistance. Rictor inhibition completely eliminated CHKA-induced enhancement of cell migration and invasion. Inhibition of mTORC1/mTORC2 reduced tumor metastasis, but the inhibition of mTORC1 alone by rapamycin exerted no effect on tumor metastasis (57). Dual inhibition of mTORC1/mTORC2 by OSI027 reversed the high expression of MDR1 in adriamycin-induced liver cancer, thereby improving the sensitivity of cancer cells to adriamycin (58).

Cholangiocarcinoma (CCC) is a highly malignant tumor. In a previous study, HCC and CCC cells were treated with sorafenib, a multikinase inhibitor of the

RAF/extracellular-signal-regulated kinase (ERK) kinase (MEK)/ERK pathway, to study the differences in signaling pathways among cell lines. Sorafenib inhibited the growth of HCC cells significantly more than that of CCC cells but minimally suppressed ERK phosphorylation. Correspondingly, sorafenib decreased Akt phosphorylation at Ser473 in HCC cells but increased Akt phosphorylation at Ser473 and mTORC2 in CCC cells. Rictor downregulation by small inhibitory RNA in RBE cells (a CCC-derived cell line) disrupted mTORC2 and inhibited Akt phosphorylation at Ser473, which promoted apoptosis and inhibited RBE cell proliferation by increasing Forkhead box protein O1. Inhibition of mTORC2 activity in the Akt/mTOR signaling pathway during sorafenib treatment to prevent the escape of the RAF/MEK/ERK pathway may lead to promising treatments for CCC (59).

Gastroesophageal cancer. The TCGA database shows that the mutation rate of Rictor in patients with esophageal GC is ~10.5%. Rictor knockdown by short hairpin RNA enhanced the inhibitory effect of LY294002 on the *in vitro* proliferation, migration and colony formation of ECa109 and EC9706 cells, which also caused cell cycle arrest and apoptosis in these cells. Furthermore, stable knockdown of Rictor *in vivo* enhanced the antitumor effect of LY294002 by promoting apoptosis and inhibiting tumor growth (60). A previous study identified 70.6% (142/201) Rictor positivity in ESCC samples (14). Furthermore, the American Joint Committee on Cancer staging was found to be positively correlated with Rictor expression and negatively associated with survival. mTOR overexpression is common in GC. Wang *et al* (33) found that Rictor protein overexpression and Rictor and *Helicobacter pylori* status may have a prognostic role in GC. A previous study by our group showed that Rictor inhibits apoptosis of GC cells by activating Cav1 through the Akt signaling pathway (29). Seo *et al* (18) reported that miR-let-7A regulates autophagy by targeting Rictor in GC cells. In other words, Rictor is involved in the autophagy of GC cells. Bian *et al* (7) analyzed 396 GC tissue samples and found that patients who were positive for Rictor and p-Akt (Ser473) expression had lower overall and relapse-free survival rates than those negative for Rictor expression. Rictor amplification is also related to tumor size, invasion depth, tumor thrombosis and tumor stage. In line with these observations, another study showed that targeting Rictor inhibited the proliferation and promoted the apoptosis of GC cells (12). Furthermore, Kim *et al* (61) reported that AZD2014, a dual mTORC1/2 inhibitor, significantly inhibited the proliferation of a Rictor-amplified patient-derived cell (PDC) line. Rictor knockdown can reverse the sensitivity of AZD2014 to PDCs. These results supported the need for further preclinical and clinical investigations with AZD2014 in Rictor-amplified GC and highlighted the importance of genomic profiling.

Pancreatic cancer. Pancreatic cancer is a devastating disease with the worst outcomes among human cancers (62). Rictor blockers reportedly inhibit tumor growth by reducing AGC kinase activation and hypoxia-inducible factor 1- α and VEGF-A expression (63). Everolimus, a Food and Drug Administration-approved mTOR inhibitor, can act in

conjunction with KPT-9274, a dual inhibitor of p21-activated kinase 4 (PAK4)-nicotinamide phosphoribosyltransferase. *In vitro* synergy with everolimus was supported by mTORC2 modification through the downregulation of Rictor, as revealed by molecular analysis. By inhibiting PAK4, KPT-9274 reduced β -catenin activity, indicating the interaction between Rho GTPases and Wnt signalling in metastatic pancreatic neuroendocrine tumors (64). Zhao *et al* (65) found that mTORC1 and mTORC2 have dual but not redundant regulatory roles in acinar-to-ductal metaplasia and early pancreatic cancer by promoting the function of the actin-related protein 2/3 (ARP2/3) complex. The ARP2/3 complex, as a co-effector of mTORC1 and mTORC2, bridges the gap between oncogenic signaling and actin dynamics of pancreatic ductal adenocarcinoma initiation. In addition, miR-155 exacerbates impaired autophagy in pancreatic acinar cells treated with caerulein by targeting Rictor (66). Gemcitabine in combination with the pro-apoptotic cytokine TNF-related apoptosis-inducing ligand inhibited the survival and induced apoptosis of pancreatic cancer cells. This combination therapy significantly increased the levels of the low phosphorylated form of tumor suppressor protein 4E-BP1. This phenomenon can be attributed to the mTOR inhibition resulting from the caspase-mediated cleavage of the Raptor and Rictor components of mTOR (67).

5. Targeted therapy

A previous study showed that rapamycin, a first-generation mTOR inhibitor, is significantly less toxic than other drugs in the effective dose range (68). The protective effect of 5-fluorouracil-rapamycin-cyclophosphamide sequential therapy is stronger than that of 5-fluorouracil-adriamycin-cyclophosphamide sequential therapy in 38 mice with colon tumors. Rapamycin has been administered for tumor treatment, but an increasing number of studies have confirmed that it was not as successful as expected in clinical trials, likely due to two reasons: First, mTOR complexes have different degrees of sensitivity to rapamycin. Since rapamycin is sensitive to mTORC1, the drug primarily inhibits the mTORC1/S6K pathway and lowers IGF-1R, which then activates mTORC2 to activate Akt (45). The activation of Akt promotes cell survival and drug resistance; thus, mTORC1 inhibitor therapy may not be beneficial. Furthermore, inhibition of mTORC2 may eliminate the adverse signaling effects of mTOR1 inhibitors. Further studies are warranted to identify potential therapeutic targets of mTORC2 and explore its related molecular mechanisms in tumors (69-71).

The results of multiple clinical trials showed that second-generation 'rapalogs' possess effective pharmacokinetic properties and exert anticancer effects (72). Table I provides a list of different types of mTORC2 inhibitors to treat CRC (73-84), liver cancer (85-99), gallbladder cancer (100-102), GC (61,103-106), esophageal cancer (32,107-110), pancreatic cancer (111-119) and biliary tract cancer (120-123). The therapeutic efficacy of rapalogs may be diminished by the pro-survival feedback loops that may be induced by the rapalogs' mTORC1-specific inhibition, such as the PI3K-Akt and PI3K-RAS-ERK pathways. Therefore, some of the drawbacks of rapalogs may be

resolved and a higher antitumor activity may be achieved by combination therapy or through the use of second-generation mTOR inhibitors, such as dual mTOR/PI3K (124) and selective mTORC1/2 inhibitors (125). However, no particular mTORC2 inhibitor has so far been identified. Therefore, it is critical to discover a specific medication that blocks Rictor. According to an *in vitro* study, CID613034 prevents the phosphorylation of mTORC2 substrates, such as Akt (Ser-473), N-myc downstream-regulated gene 1 (TR-346) and PKC; however, the phosphorylation state of the mTORC1 substrate S6K (Thr-389) or the mTORC1-dependent negative feedback loop are unaffected (126).

According to Werfel *et al* (127), RNAi therapy based on nanoparticles successfully silences Rictor. Through the intratumoral and intravenous delivery of nanoparticle-based Rictor, tumor-cell inhibition and Akt phosphorylation were observed to be decreased in HER2-amplified breast cancers. HER2-amplified breast cancer is less likely to spread when selective mTORC2 inhibitor therapy is paired with the HER2 inhibitor lapatinib. This suggests that mTORC2 encourages lapatinib resistance. The potential for beneficial anticancer effects of mTOR inhibitors in transplant-associated malignancies and other cancers is also highlighted by preclinical and clinical findings (128). Preclinical research has demonstrated that Rictor controls the biological activities of different immune cells, and that its knock-down improves the effectiveness of immunotherapy (129). Targeting mTOR in immune cells, however, has the potential to impair immunological tolerance and cause autoimmune disorders (130,131). Failure of immunotherapy is frequently caused by autoimmune diseases. A list of clinical trials on Rictor inhibitors in gastrointestinal cancer is provided in Table II (132-136). At present, second-generation mTOR inhibitors include dual mTOR/PI3K inhibitors, such as PI-103, NVP-BEZ235 and WJD008; selective mTORC1/2 inhibitors, such as Torin1, PP242 and PP30, and others, such as Ku-0063794, WAY-600, WYE-687 and WYE354, which have been reported to be ATP-competitive mTOR inhibitors, as they effectively inhibit mTORC1 and mTORC2 (137). Most of these drugs are in clinical trials and available data suggests that combination regimens are better than monotherapies.

6. Conclusion and future perspectives

Rictor, a key effector of the PI3K/Akt/mTOR pathway, has an important role in tumor development and invasion. It causes tumor resistance through Akt-dependent and -independent pathways, severely limiting the efficacy of targeted drugs. Therefore, Rictor is an important potential target for addressing drug resistance.

Rictor/mTORC2 alterations are more frequent in a variety of tumor types. However, the mechanisms of Rictor/oncogenic mTORC2 remain to be further clarified. It is essential to understand how Rictor/oncogenic mTORC2 relates to other PI3K/mTOR signalling pathways. Currently available Rictor/mTORC2 inhibitors are second-generation mTOR inhibitors, and their inhibitory effects on Rictor/mTORC2 comprise dual mTOR/PI3K, selective mTORC1/2 inhibition and ATP-competitive mTOR inhibition. The impacts of

Table I. Different types of mTORC2 inhibitor.

Cancer/cell type	Drug classification			(Refs.)
	1	2	3	
Colorectal cancer	Dactolisib (BEZ235) MM-129 LY3023414	OSI-207 Zotarolimus PP242 WYE-354	AZD8055 AZD2014	(73-84)
Liver cancer	NVP-BEZ235	CC-223 Ku0063794 OSI-207 RAD001 FK506	MLN0128 (NK128) AZD8055 AZD2014	(85-99)
Gallbladder cancer	-	WYE-354 OSI-207 RAD001	-	(100-102)
Gastric cancer	NVP-BEZ235	RAD001 OSI-207 PP242	AZD2014	(61,103-106)
Esophageal cancer	LY3023414 NVP-BEZ235	PP242	-	(32,107-110)
Pancreatic cancer	-	OSI-207 PP242 RAD001	AZD8055 MLN0128 (NK128) AZD2014	(111-119)
Biliary tract cancer	LY3023414	OSI-207 RAD001	-	(120-123)

Drug classes: 1, mTOR and PI3K dual specificity inhibitors; 2, selective mammalian target of rapamycin complex 1/2 inhibitors; 3, others.

Table II. Preclinical evaluation of Rictor/mTOR complex 2 inhibitors in gastrointestinal tumors.

Cancer type	Rictor inhibitor	NCI number	Clinical phase	Country and region	(Refs.)
Colorectal cancer	LY3023414	NCT02124148	I	North America: US	(132)
Liver cancer	CC223 RAD001	NCT03591965 NCT01035229	II III	Asia: China World: East Asia, Europe, Middle East, Pacifica, North America, Southeast Asia	Recruiting (133)
	FK506	NCT00355862	III	Europe	(134)
Gallbladder cancer	-	-	-	-	-
Gastric cancer	RAD001	NCT01231399	I/II	North America: US	(135)
Esophageal cancer	RAD001	NCT01231399	I/II	North America: US	(135)
Pancreatic cancer	RAD001	NCT00560963	I/II	Europe	(136)
Biliary tract cancer	-	-	-	-	-

the second-generation mTOR inhibitors on gastrointestinal cancers showed better treatment efficacy than monotherapies in *in vitro* cell experiments and preclinical studies. However, no special inhibitors of Rictor/mTORC2 have been identified (138).

At present, PI3K/mTOR inhibitors cannot serve as effective treatment agents. The therapeutic benefits of select molecular inhibitors may be useful if patients are classified based on their Rictor alteration status.

Acknowledgements

Not applicable.

Funding

No funding was received.

Availability of data and materials

Not applicable.

Authors' contributions

All authors contributed to the article and have read and approved the submitted version. RC and LM reviewed the literature and collated and analysed the information. SG and

PL conceived and designed the study. RC, SG and PL drafted the manuscript. Data authentication is not applicable.

Ethics approval and consent to participate

Not applicable.

Patient consent for publication

Not applicable.

Competing interests

The authors declare they have no competing interests.

References

- Sung H, Ferlay J, Siegel RL, Laversanne M, Soerjomataram I, Jemal A and Bray F: Global Cancer Statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin* 71: 209-249, 2021.
- Yang L, Ying X, Liu S, Lyu G, Xu Z, Zhang X, Li H, Li Q, Wang N and Ji J: Gastric cancer: Epidemiology, risk factors and prevention strategies. *Chin J Cancer Res* 32: 695-704, 2020.
- Lu L, Mullins CS, Schafmayer C, Zeißig S and Linnebacher M: A global assessment of recent trends in gastrointestinal cancer and lifestyle-associated risk factors. *Cancer Commun (Lond)* 41: 1137-1151, 2021.
- Hou J, He Z, Liu T, Chen D, Wang B, Wen Q and Zheng X: Evolution of molecular targeted cancer therapy: Mechanisms of drug resistance and novel opportunities identified by CRISPR-Cas9 Screening. *Front Oncol* 12: 755053, 2022.
- Zhao D, Jiang M, Zhang X and Hou H: The role of Rictor amplification in targeted therapy and drug resistance. *Mol Med* 26: 20, 2020.
- Cerami E, Gao J, Dogrusoz U, Gross BE, Sumer SO, Aksoy BA, Jacobsen A, Byrne CJ, Heuer ML, Larsson E, *et al*: The cBio cancer genomics portal: An open platform for exploring multidimensional cancer genomics data. *Cancer Discov* 2: 401-404, 2012.
- Bian Y, Wang Z, Xu J, Zhao W, Cao H and Zhang Z: Elevated Rictor expression is associated with tumor progression and poor prognosis in patients with gastric cancer. *Biochem Biophys Res Commun* 464: 534-540, 2015.
- Wang LF, Chen HJ, Yu JL, Qi J, Lin XH and Zou ZW: Expression of Rictor and mTOR in colorectal cancer and their clinical significance. *Nan Fang Yi Ke Da Xue Xue Bao* 36: 396-400, 2016 (In Chinese).
- Jiang WJ, Feng RX, Liu JT, Fan LL, Wang H and Sun GP: RICTOR expression in esophageal squamous cell carcinoma and its clinical significance. *Med Oncol* 34: 32, 2017.
- Beauchamp EM and Platanias LC: The evolution of the TOR pathway and its role in cancer. *Oncogene* 32: 3923-3932, 2013.
- Murugan AK: mTOR: Role in cancer, metastasis and drug resistance. *Semin Cancer Biol* 59: 92-111, 2019.
- Gaubitz C, Prouteau M, Kusmider B and Loewith R: TORC2 structure and function. *Trends Biochem Sci* 41: 532-545, 2016.
- Sarbassov DD, Ali SM, Kim DH, Guertin DA, Latek RR, Erdjument-Bromage H, Tempst P and Sabatini DM: Rictor, a novel binding partner of mTOR, defines a rapamycin-insensitive and raptor-independent pathway that regulates the cytoskeleton. *Curr Biol* 14: 1296-1302, 2004.
- Zhou P, Zhang N, Nussinov R and Ma B: Defining the domain arrangement of the mammalian target of rapamycin complex component rictor protein. *J Comput Biol* 22: 876-886, 2015.
- Wang MC, Wu AG, Huang YZ, Shao GL, Ji SF, Wang RW, Yuan HJ, Fan XL, Zheng LH and Jiao QL: Autophagic regulation of cell growth by altered expression of Beclin 1 in triple-negative breast cancer. *Int J Clin Exp Med* 8: 7049-7058, 2015.
- Sui H, Shi C, Yan Z and Li H: Combination of erlotinib and a PARP inhibitor inhibits growth of A2780 tumor xenografts due to increased autophagy. *Drug Des Devel Ther* 9: 3183-3190, 2015.
- Fan H, Jiang M, Li B, He Y, Huang C, Luo D, Xu H, Yang L and Zhou J: MicroRNA-let-7a regulates cell autophagy by targeting Rictor in gastric cancer cell lines MGC-803 and SGC-7901. *Oncol Rep* 39: 1207-1214, 2018.
- Seo SU, Woo SM, Lee HS, Kim SH, Min KJ and Kwon TK: mTORC1/2 inhibitor and curcumin induce apoptosis through lysosomal membrane permeabilization-mediated autophagy. *Oncogene* 37: 5205-5220, 2018.
- Liu Y, Sun Y and Zhao A: MicroRNA-134 suppresses cell proliferation in gastric cancer cells via targeting of GOLPH3. *Oncol Rep* 37: 2441-2448, 2017.
- Sarbassov DD, Guertin DA, Ali SM and Sabatini DM: Phosphorylation and regulation of Akt/PKB by the rictor-mTOR complex. *Science* 307: 1098-1101, 2005.
- Hresko RC and Mueckler M: mTOR. Rictor is the Ser473 kinase for Akt/protein kinase B in 3T3-L1 adipocytes. *J Biol Chem* 280: 40406-40416, 2005.
- Yuan TL and Cantley LC: PI3K pathway alterations in cancer: Variations on a theme. *Oncogene* 27: 5497-5510, 2008.
- Treins C, Warne PH, Magnuson MA, Pende M and Downward J: Rictor is a novel target of p70 S6 kinase-1. *Oncogene* 29: 1003-1016, 2010.
- Lang F, Strutz-Seebohm N, Seebohm G and Lang UE: Significance of SGK1 in the regulation of neuronal function. *J Physiol* 588: 3349-3354, 2010.
- Leong ML, Maiyar AC, Kim B, O'Keeffe BA and Firestone GL: Expression of the serum- and glucocorticoid-inducible protein kinase, Sgk, is a cell survival response to multiple types of environmental stress stimuli in mammary epithelial cells. *J Biol Chem* 278: 5871-5882, 2003.
- García-Martínez Juan M and Alessi Dario R: mTOR complex 2 (mTORC2) controls hydrophobic motif phosphorylation and activation of serum- and glucocorticoid-induced protein kinase 1 (SGK1). *Biochem J* 416: 375-385, 2008.
- Zhao L, Zhu L, Oh YT, Qian G, Chen Z and Sun SY: Rictor, an essential component of mTOR complex 2, undergoes caspase-mediated cleavage during apoptosis induced by multiple stimuli. *Apoptosis* 26: 338-347, 2021.
- Wen FF, Li XY, Li YY, He S, Xu XY, Liu YH, Liu L and Wu SH: Expression of Raptor and Rictor and their relationships with angiogenesis in colorectal cancer. *Neoplasma* 67: 501-508, 2020.
- Cao RZ, Min L, Liu S, Tian RY, Jiang HY, Liu J, Shao LL, Cheng R, Zhu ST, Guo SL and Li P: Rictor activates Cav 1 through the Akt signaling pathway to inhibit the apoptosis of gastric cancer cells. *Front Oncol* 11: 641453, 2021.
- Liu H, Zang H, Kong J and Gong L: In vivo and impact of miRNA-153 on the suppression of cell growth apoptosis through mTORC2 signaling pathway in breast cancer. *J Recept Signal Transduct Res* 42: 390-398, 2022.
- Hou B, Liu S, Li E and Jiang X: Different role of raptor and rictor in regulating Rasfonin-Induced autophagy and apoptosis in renal carcinoma cells. *Chem Biodivers* 17: e2000743, 2020.
- Lu Z, Shi X, Gong F, Li S, Wang Y, Ren Y, Zhang M, Yu B, Li Y, Zhao W, *et al*: Rictor/mTORC2 affects tumorigenesis and therapeutic efficacy of mTOR inhibitors in esophageal squamous cell carcinoma. *Acta Pharm Sin B* 10: 1004-1019, 2020.
- Wang F, Lou X, Zou Y, Hu D, Liu J, Ning J, Jiao Y, Zhang Z, Yang F, Fan L, *et al*: Overexpression of Rictor protein and Rictor-H. pylori interaction has impact on tumor progression and prognosis in patients with gastric cancer. *Folia Histochem Cytobiol* 58: 96-107, 2020.
- Wang S, Amato KR, Song W, Youngblood V, Lee K, Boothby M, Brantley-Sieders DM and Chen J: Regulation of endothelial cell proliferation and vascular assembly through distinct mTORC2 signaling pathways. *Mol Cell Biol* 35: 1299-1313, 2015.
- Liang X, Sun R, Zhao X, Zhang Y, Gu Q, Dong X, Zhang D, Sun J and Sun B: Rictor regulates the vasculogenic mimicry of melanoma via the Akt-MMP-2/9 pathway. *J Cell Mol Med* 21: 3579-3591, 2017.
- Guan B, Wu K, Zeng J, Xu S, Mu L, Gao Y, Wang K, Ma Z, Tian J, Shi Q, *et al*: Tumor-suppressive microRNA-218 inhibits tumor angiogenesis via targeting the mTOR component Rictor in prostate cancer. *Oncotarget* 8: 8162-8172, 2027.
- Dormond O, Contreras AG, Meijer E, Datta D and Flynn E: CD40-induced signaling in human endothelial cells results in mTORC2- and Akt-dependent expression of vascular endothelial growth factor in vitro and in vivo. *J Immunol* 181: 8088-8095, 2008.
- Alizadeh AM, Shiri S and Farsinejad S: Metastasis review: From bench to bedside. *Tumour Biol* 35: 8483-8523, 2014.
- Guertin DA, Stevens DM, Thoreen CC, Burds AA, Kalaany NY, Moffat J, Brown M, Fitzgerald KJ and Sabatini DM: 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* 11: 859-871, 2006.

40. Hua H, Kong Q, Zhang H, Wang J, Luo T and Jiang Y: Targeting mTOR for cancer therapy. *J Hematol Oncol* 12: 71, 2019.
41. Agarwal NK, Chen CH, Cho H, Boulbès DR, Spooner E and Sarbassov DD: Rictor regulates cell migration by suppressing RhoGDI2. *Oncogene* 32: 2521-2526, 2013.
42. Agarwal NK, Kazyken D and Sarbassov dos D: Rictor encounters RhoGDI2: The second pilot is taking a lead. *Small GTPases* 4: 102-105, 2013.
43. Savukaitytė A, Gudoitytė G, Bartnykaitė A, Ugenskienė R and Juozaitytė E: siRNA knockdown of REDD1 facilitates aspirin-mediated dephosphorylation of mTORC1 target 4E-BP1 in MDA-MB-468 human breast cancer cell line. *Cancer Manag Res* 13: 1123-1133, 2021.
44. Wei F, Zhang Y, Geng L, Zhang P, Wang G and Liu Y: mTOR inhibition induces EGFR feedback activation in association with its resistance to human pancreatic cancer. *Int J Mol Sci* 16: 3267-3282, 2015.
45. Lang SA, Hackl C, Moser C, Fichtner-Feigl S, Koehl GE, Schlitt HJ, Geissler EK and Stoeltzing O: Implication of Rictor in the mTOR inhibitor-mediated induction of insulin-like growth factor-I receptor (IGF-IR) and human epidermal growth factor receptor-2 (HER2) expression in gastrointestinal cancer cells. *Biochim Biophys Acta* 1803: 435-442, 2010.
46. Yin Y, Hua H, Li M, Liu S, Kong Q, Shao T, Wang J, Luo Y, Wang Q, Luo T, *et al*: mTORC2 promotes type I insulin-like growth factor receptor and insulin receptor activation through the tyrosine kinase activity of Mtor. *Cell Res* 26: 46-65, 2016.
47. Hossain MS, Karuniawati H, Jairoun AA, Urbi Z, Ooi J, John A, Lim YC, Kibria KMK, Mohiuddin AKM, Ming LC, *et al*: Colorectal cancer: A review of carcinogenesis, global epidemiology, current challenges, risk factors, preventive and treatment strategies. *Cancers (Basel)* 14: 1732, 2022.
48. Bellier J, Nokin MJ, Caprasse M, Tiamiou A, Blomme A, Scheijen JL, Koopmansch B, MacKay GM, Chiavarina B, Costanza B, *et al*: Methylglyoxal scavengers resensitize KRAS-Mutated colorectal tumors to cetuximab. *Cell Rep* 30: 1400-1416.e6, 2020.
49. Shuhua W, Chenbo S, Yangyang L, Xiangqian G, Shuang H, Tangyue L and Dong T: Autophagy-related genes Raptor, Rictor, and Beclin 1 expression and relationship with multidrug resistance in colorectal carcinoma. *Hum Pathol* 46: 1752-1759, 2015.
50. Wei Y, Tang X, Ren Y, Yang Y, Song F, Fu J, Liu S, Yu M, Chen J, Wang S, *et al*: An RNA-RNA crosstalk network involving HMGB1 and RICTOR facilitates hepatocellular carcinoma tumorigenesis by promoting glutamine metabolism and impedes immunotherapy by PD-L1+ exosomes activity. *Signal Transduct Target Ther* 6: 421, 2021.
51. Reyes-Gordillo K, Shah R, Arellanes-Robledo J, Cheng Y, Ibrahim J and Tuma PL: Akt1 and Akt2 isoforms play distinct roles in regulating the development of inflammation and fibrosis associated with alcoholic liver disease. *Cells* 8: 1337, 2019.
52. Guri Y, Colombi M, Dazert E, Hindupur SK, Roszik J, Moes S, Jenoe P, Heim MH, Riezman I, Riezman H and Hall MN: mTORC2 promotes tumorigenesis via lipid synthesis. *Cancer Cell* 32: 807-823.e12, 2017.
53. Dong X, Feng M, Yang H, Liu H, Guo H, Gao X, Liu Y, Liu R, Zhang N, Chen R and Kong R: Rictor promotes cell migration and actin polymerization through regulating ABLIM1 phosphorylation in Hepatocellular Carcinoma. *Int J Biol Sci* 16: 2835-2852, 2020.
54. Hu J, Che L, Li L, Pilo MG, Cigliano A, Ribback S, Li X, Latte G, Mela M, Evert M, *et al*: Co-activation of Akt and c-Met triggers rapid hepatocellular carcinoma development via the mTORC1/FASN pathway in mice. *Sci Rep* 6: 20484, 2016.
55. Villanueva A, Chiang DY, Newell P, Peix J, Thung S, Alsinet C, Tovar V, Roayaie S, Minguez B, Sole M, *et al*: Pivotal role of mTOR signaling in hepatocellular carcinoma. *Gastroenterology* 135: 1972-1983, 1983.e1-e11, 2008.
56. Xu Z, Xu M, Liu P, Zhang S, Shang R, Qiao Y, Che L, Ribback S, Cigliano A, Evert K, *et al*: The mTORC2-Akt1 Cascade Is Crucial for c-Myc to Promote Hepatocarcinogenesis in Mice and Humans. *Hepatology* 70: 1600-1613, 2019.
57. Lin XM, Hu L, Gu J, Wang RY, Li L, Tang J, Zhang BH, Yan XZ, Zhu YJ, Hu CL, *et al*: Choline Kinase α mediates interactions between the epidermal growth factor receptor and mechanistic target of rapamycin complex 2 in hepatocellular carcinoma cells to promote drug resistance and xenograft tumor progression. *Gastroenterology* 152: 1187-1202, 2017.
58. Joechle K, Guenzle J, Hellerbrand C, Strnad P, Cramer T, Neumann UP and Lang SA: Role of mammalian target of rapamycin complex 2 in primary and secondary liver cancer. *World J Gastrointest Oncol* 13: 1632-1647, 2021.
59. Yokoi K, Kobayashi A, Motoyama H, Kitazawa M, Shimizu A, Notake T, Yokoyama T, Matsumura T, Takeoka M and Miyagawa SI: Survival pathway of cholangiocarcinoma via Akt/mTOR signaling to escape RAF/MEK/ERK pathway inhibition by sorafenib. *Oncol Rep* 39: 843-850, 2018.
60. Hou G, Zhao Q, Zhang M, Fan T, Liu M, Shi X, Ren Y, Wang Y, Zhou J and Lu Z: Down-regulation of Rictor enhances cell sensitivity to PI3K inhibitor LY294002 by blocking mTORC2-mediated phosphorylation of Akt/PRAS40 in esophageal squamous cell carcinoma. *Biomed Pharmacother* 106: 1348-1356, 2018.
61. Kim ST, Kim SY, Klempner SJ, Yoon J, Kim N, Ahn S, Bang H, Kim KM, Park W, Park SH, *et al*: Rapamycin-insensitive companion of mTOR (RICTOR) amplification defines a subset of advanced gastric cancer and is sensitive to AZD2014-mediated mTORC1/2 inhibition. *Ann Oncol* 28: 547-554, 2017.
62. Siegel RL, Miller KD and Jemal A: Cancer statistics, 2019. *CA Cancer J Clin* 69: 7-34, 2019.
63. Schmidt KM, Hellerbrand C, Ruemmele P, Michalski CW, Kong B, Kroemer A, Hackl C, Schlitt HJ, Geissler EK and Lang SA: Inhibition of mTORC2 component Rictor impairs tumor growth in pancreatic cancer models. *Oncotarget* 8: 24491-24505, 2017.
64. Mpilla GB, Uddin MH, Al-Hallak MN, Aboukameel A, Li Y, Kim SH, Beydoun R, Dyson G, Baloglu E, Senapedis WT, *et al*: PAK4-NAMPT dual inhibition sensitizes pancreatic neuroendocrine tumors to everolimus. *Mol Cancer Ther* 20: 1836-1845, 2021.
65. Zhao Y, Schoeps B, Yao D, Zhang Z, Schuck K, Tissen V, Jäger C, Schlitter AM, van der Kammen R, Ludwig C, *et al*: mTORC1 and mTORC2 Converge on the Arp2/3 complex to promote Kras-induced Acinar-to-ductal metaplasia and early pancreatic carcinogenesis. *Gastroenterology* 160: 1755-1770.e17, 2021.
66. Zhang X, Chu J, Sun H, Zhao D, Ma B, Xue D, Zhang W and Li Z: MiR-155 aggravates impaired autophagy of pancreatic acinar cells through targeting Rictor. *Acta Biochim Biophys Sin (Shanghai)* 52: 192-199, 2020.
67. Elia A, Henry-Grant R, Adiseshiah C, Marboeuf C, Buckley RJ, Clemens MJ, Mudan S and Pyronnet S: Implication of 4E-BP1 protein dephosphorylation and accumulation in pancreatic cancer cell death induced by combined gemcitabine and TRAIL. *Cell Death Dis* 8: 3204, 2017.
68. Eng CP, Sehgal SN and Vézina C: Activity of rapamycin (AY-22,989) against transplanted tumors. *J Antibiot (Tokyo)* 37: 1231-1237, 1984.
69. Chiarini F, Evangelisti C, McCubrey JA and Martelli AM: Current treatment strategies for inhibiting Mtor in cancer. *Trends Pharmacol Sci* 36: 124-35, 2015.
70. Wu SH, Bi JF, Cloughesy T, Cavenee WK and Mischel PS: Emerging function of mTORC2 as a core regulator in Glioblastoma: Metabolic reprogramming and drug resistance. *Cancer Biol Med* 11: 255-263, 2014.
71. Masui K, Harachi M, Cavenee WK, Mischel PS and Shibata N: mTOR Complex 2 is an integrator of cancer metabolism and epigenetics. *Cancer Lett* 478: 1-7, 2020.
72. Zhou HY and Huang SL: Current development of the second generation of mTOR inhibitors as anticancer agents. *Chin J Cancer* 31: 8-18, 2012.
73. Hu Y, Zhang K, Zhu X, Zheng X, Wang C, Niu X, Jiang T, Ji X, Zhao W, Pang L, *et al*: Synergistic inhibition of drug-resistant colon cancer growth with PI3K/mTOR dual inhibitor BEZ235 and Nano-emulsified paclitaxel via reducing multidrug resistance and promoting apoptosis. *Int J Nanomedicine* 16: 2173-2186, 2021.
74. Hermanowicz JM, Kalaska B, Pawlak K, Sieklucka B, Miklosz J, Mojzych M and Pawlak D: Preclinical toxicity and safety of MM-129-First-in-Class BTK/PD-L1 inhibitor as a potential candidate against colon cancer. *Pharmaceutics* 13: 1222, 2021.
75. Foley TM, Payne SN, Pasch CA, Yueh AE, Van De Hey DR, Korkos DP, Clipson L, Maher ME, Matkowskyj KA, Newton MA and Deming DA: APC dual PI3K/mTOR inhibition in colorectal cancers with and mutations. *Mol Cancer Res* 15: 317-327, 2017.
76. Lou J, Lv JX, Zhang YP and Liu ZJ: OSI-027 inhibits the tumorigenesis of colon cancer through mediation of c-Myc/FOXO3a/PUMA axis. *Cell Biol Int* 46: 1204-1214, 2022.

77. Wang H, Liu Y, Ding J, Huang Y, Liu J, Liu N, Ao Y, Hong Y, Wang L, Zhang L, *et al*: Targeting mTOR suppressed colon cancer growth through 4EBP1/eIF4E/PUMA pathway. *Cancer Gene Ther* 27: 448-460, 2020.
78. Chang GR, Kuo CY, Tsai MY, Lin WL, Lin TC, Liao HJ, Chen CH and Wang YC: Anti-cancer effects of zotarolimus combined with 5-fluorouracil treatment in HCT-116 colorectal cancer-bearing BALB/c Nude Mice. *Molecules* 26: 4683, 2021.
79. Rashid MM, Lee H and Jung BH: Evaluation of the antitumor effects of PP242 in a colon cancer xenograft mouse model using comprehensive metabolomics and lipidomics. *Sci Rep* 10: 17523, 2020.
80. Wang L, Zhu YR, Wang S and Zhao S: Autophagy inhibition sensitizes WYE-354-induced anti-colon cancer activity in vitro and in vivo. *Tumor Biol* 37: 11743-11752, 2016.
81. Chen Y, Lee CH, Tseng BY, Tsai YH, Tsai HW, Yao CL and Tseng SH: AZD8055 exerts antitumor effects on colon cancer cells by inhibiting mTOR and Cell-cycle Progression. *Anticancer Res* 38: 1445-1454, 2018.
82. Jin ZZ, Wang W, Fang DL and Jin YJ: mTOR inhibition sensitizes ONC201-induced anti-colorectal cancer cell activity. *Biochem Biophys Res Commun* 478: 1515-1520, 2016.
83. Nguyen DQ, Hoang DH, Nelson M, Nigam L, Nguyen VTT, Zhang L, Pham TKT, Ho HD, Nguyen DDT, Lam TQ, *et al*: Requirement of GTP binding for TIF-90-regulated ribosomal RNA synthesis and oncogenic activities in human colon cancer cells. *J Cell Physiol* 235: 7567-7579, 2020.
84. Reita D, Bour C, Benbrika R, Groh A, Pencreach E, Guérin E and Guenot D: Synergistic Anti-tumor effect of mTOR inhibitors with irinotecan on colon cancer cells. *Cancers (Basel)* 11: 1581, 2019.
85. Wang Y, Miao X, Jiang Y, Wu Z, Zhu X, Liu H, Wu X, Cai J, Ding X and Gong W: The synergistic antitumor effect of IL-6 neutralization with NVP-BEZ235 in hepatocellular carcinoma. *Cell Death Dis* 13: 146, 2022.
86. Narahara S, Watanabe T, Nagaoka K, Fujimoto N, Furuta Y, Tanaka K, Tokunaga T, Kawasaki T, Yoshimaru Y, Setoyama H, *et al*: Clusterin and related scoring index as potential early predictors of response to sorafenib in hepatocellular carcinoma. *Hepatol Commun* 6: 1198-1212, 2022.
87. Cao W, Liu X, Zhang Y, Li A, Xie Y, Zhou S, Song L, Xu R, Ma Y, Cai S and Tang X: BEZ235 increases the sensitivity of hepatocellular carcinoma to sorafenib by inhibiting PI3K/Akt/mTOR and inducing autophagy. *Biomed Res Int* 2021: 5556306, 2021.
88. Liang Y, Xie C, Li A, Huo Z, Wu B, Cai S, Cao W, Ma Y, Xu R, Jiang Z, *et al*: Anti-GPC3 Antibody-Conjugated BEZ235 loaded polymeric nanoparticles (Ab-BEZ235-NP) enhances radiosensitivity in hepatocellular carcinoma cells by inhibition of DNA double-strand break repair. *J Biomed Nanotechnol* 16: 446-455, 2020.
89. Xie Z, Wang J, Liu M, Chen D, Qiu C and Sun K: CC-223 blocks mTORC1/C2 activation and inhibits human hepatocellular carcinoma cells in vitro and in vivo. *PLoS One* 12: e0173252, 2017.
90. Choi HJ, Park JH, Kim OH, Kim KH, Hong HE, Seo H and Kim SJ: Combining Everolimus and Ku0063794 Promotes apoptosis of hepatocellular carcinoma cells via reduced autophagy resulting from diminished expression of miR-4790-3p. *Int J Mol Sci* 22: 2859, 2021.
91. Yongxi T, Haijun H, Jiaping Z, Guoliang S and Hongying P: Autophagy inhibition sensitizes KU-0063794-mediated anti-HepG2 hepatocellular carcinoma cell activity in vitro and in vivo. *Biochem Biophys Res Commun* 465: 494-500, 2015.
92. Zhen MC, Wang FQ, Wu SF, Zhao YL, Liu PG and Yin ZY: Identification of mTOR as a primary resistance factor of the IAP antagonist AT406 in hepatocellular carcinoma cells. *Oncotarget* 8: 9466-9475, 2017.
93. Kaneya Y, Takata H, Wada R, Kure S, Ishino K, Kudo M, Kondo R, Taniai N, Ohashi R, Yoshida H and Naito Z: Inhibitor for protein disulfide-isomerase family A member 3 enhances the antiproliferative effect of inhibitor for mechanistic target of rapamycin in liver cancer: An study on combination treatment with everolimus and 16F16. *Oncol Lett* 21: 28, 2021.
94. Navarro-Villarán E, de la Cruz-Ojeda P, Contreras L, González R, Negrete M, Rodríguez-Hernández MA, Marín-Gómez LM, Álamo-Martínez JM, Calvo A, Gómez-Bravo MA, *et al*: Molecular pathways leading to induction of cell death and anti-proliferative properties by tacrolimus and mTOR inhibitors in liver cancer cells. *Cell Physiol Biochem* 54: 457-473, 2020.
95. Zhang S, Song X, Cao D, Xu Z, Fan B, Che L, Hu J, Chen B, Dong M, Pilo MG, *et al*: Pan-mTOR inhibitor MLN0128 is effective against intrahepatic cholangiocarcinoma in mice. *J Hepatol* 67: 1194-1203, 2017.
96. Lee HY, Lee YG, Lee S, Elvira R, Seo HE, Lee JY, Han J and Lee K: Activation of ERK and p38 reduces AZD8055-mediated inhibition of protein synthesis in hepatocellular carcinoma HepG2 cell line. *Int J Mol Sci* 22: 11824, 2021.
97. Patra T, Meyer K, Ray RB, Kanda T and Ray R: Akt inhibitor augments anti-proliferative efficacy of a dual mTORC1/2 inhibitor by FOXO3a activation in p53 mutated hepatocarcinoma cells. *Cell Death Dis* 12: 1073, 2021.
98. Liu M, Gu P, Guo W and Fan X: C6 ceramide sensitizes the anti-hepatocellular carcinoma (HCC) activity by AZD-8055, a novel mTORC1/2 dual inhibitor. *Tumor Biol* 37: 11039-11048, 2016.
99. Peng X, Zhang D, Li Z, Fu M and Liu H: mTOR inhibition sensitizes human hepatocellular carcinoma cells to resminostat. *Biochem Biophys Res Commun* 477: 556-562, 2016.
100. Weber H, Leal P, Stein S, Kunkel H, García P, Bizama C, Espinoza JA, Riquelme I, Nervi B, Araya JC, *et al*: Rapamycin and WYE-354 suppress human gallbladder cancer xenografts in mice. *Oncotarget* 6: 31877-31888, 2015.
101. Li Q, Mou LJ, Tao L, Chen W, Sun XT, Xia XF, Wu XY and Shi XL: Inhibition of mTOR suppresses human gallbladder carcinoma cell proliferation and enhances the cytotoxicity of 5-fluorouracil by downregulating MDR1 expression. *Eur Rev Med Pharmacol Sci* 20: 1699-1706, 2016.
102. Mohri D, Ijichi H, Miyabayashi K, Takahashi R, Kudo Y, Sasaki T, Asaoka Y, Tanaka Y, Ikenoue T, Tateishi K, *et al*: A potent therapeutics for gallbladder cancer by combinatorial inhibition of the MAPK and mTOR signaling networks. *J Gastroenterol* 51: 711-721, 2016.
103. Yokoyama D, Hisamori S, Deguchi Y, Nishigori T, Okabe H, Kanaya S, Manaka D, Kadokawa Y, Hata H, Minamiguchi S, *et al*: PTEN is a predictive biomarker of trastuzumab resistance and prognostic factor in HER2-overexpressing gastroesophageal adenocarcinoma. *Sci Rep* 11: 9013, 2021.
104. Gao F, Li R, Wei PF, Ou L, Li M, Bai Y, Luo WJ and Fan Z: Synergistic anticancer effects of everolimus (RAD001) and Rheon on gastric cancer cells via phosphoinositide-3-kinase (PI3K)/protein kinase B (Akt)/mammalian target of rapamycin (mTOR) pathway. *Bioengineered* 13: 6332-6342, 2022.
105. Xu E, Zhu H, Wang F, Miao J, Du S, Zheng C, Wang X, Li Z, Xu F, Xia X and Guan W: OSI-027 alleviates Oxaliplatin Chemoresistance in gastric cancer cells by suppressing P-gp induction. *Curr Mol Med* 21: 922-930, 2021.
106. Xing X, Zhang L, Wen X, Wang X, Cheng X, Du H, Hu Y, Li L, Dong B, Li Z and Ji J: PP242 suppresses cell proliferation, metastasis, and angiogenesis of gastric cancer through inhibition of the PI3K/Akt/mTOR pathway. *Anticancer Drugs* 25: 1129-1140, 2014.
107. Zaidi AH, Kosovec JE, Matsui D, Omstead AN, Raj M, Rao RR, Biederman RWW, Finley GG, Landreneau RJ, Kelly RJ and Jobe BA: PI3K/mTOR dual inhibitor, LY3023414, demonstrates potent antitumor efficacy against esophageal adenocarcinoma in a rat model. *Ann Surg* 266: 91-98, 2017.
108. Du W, Gao A, Herman JG, Wang L, Zhang L, Jiao S and Guo M: Methylation of NRN1 is a novel synthetic lethal marker of PI3K-Akt-mTOR and ATR inhibitors in esophageal cancer. *Cancer Sci* 112: 2870-2883, 2021.
109. Hou H, Zhao H, Yu X, Cong P, Zhou Y, Jiang Y and Cheng Y: METTL3 promotes the proliferation and invasion of esophageal cancer cells partly through Akt signaling pathway. *Pathol Res Pract* 216: 153087, 2020.
110. Lu Z, Zhang Y, Xu Y, Wei H, Zhao W, Wang P, Li Y and Hou G: mTOR inhibitor PP242 increases antitumor activity of sulforaphane by blocking Akt/mTOR pathway in esophageal squamous cell carcinoma. *Mol Biol Rep* 49: 451-461, 2022.
111. Chen B, Xu M, Zhang H, Xu MZ, Wang XJ, Tang QH and Tang JY: The Antipneumatic cancer activity of OSI-027, a potent and selective inhibitor of mTORC1 and mTORC2. *DNA Cell Biol* 34: 610-617, 2015.
112. Huang B, Wang J, Chen Q, Qu C, Zhang J, Chen E, Zhang Y, Wang Y, Ni L and Liang T: Gemcitabine enhances OSI-027 cytotoxicity by upregulation of miR-663a in pancreatic ductal adenocarcinoma cells. *Am J Transl Res* 11: 473-485, 2019.
113. Zhi X, Chen W, Xue F, Liang C, Chen BW, Zhou Y, Wen L, Hu L, Shen J, Bai X and Liang T: OSI-027 inhibits pancreatic ductal adenocarcinoma cell proliferation and enhances the therapeutic effect of gemcitabine both in vitro and in vivo. *Oncotarget* 6: 26230-26241, 2015.

114. Soares HP, Ni Y, Kisfalvi K, Sinnett-Smith J and Rozengurt E: Different patterns of Akt and ERK feedback activation in response to rapamycin, active-site mTOR inhibitors and metformin in pancreatic cancer cells. *PLoS One* 8: e57289, 2013.
115. Peng T and Dou QP: Everolimus inhibits growth of gemcitabine-resistant pancreatic cancer cells via induction of caspase-dependent apoptosis and G₂/M arrest. *J Cell Biochem* 118: 2722-2730, 2017.
116. Hofmann BT, Picksak AS, Kwiatkowski M, Grupp K, Jücker M, Bachmann K, Mercanoglu B, Izbicki JR, Kahlert C, Bockhorn M, *et al*: Truncated O-GalNAc glycans impact on fundamental signaling pathways in pancreatic cancer. *Glycobiology*: Aug 18, 2021 (Epub ahead of print). doi: 10.1093/glycob/cwab088.
117. Zhu J, Lv J, Chen J, Zhang X and Ji Y: Down-regulated microRNA-223 or elevated ZIC1 inhibits the development of pancreatic cancer via inhibiting PI3K/Akt/mTOR signaling pathway activation. *Cell Cycle* 19: 2851-2865, 2020.
118. Lewis CS, Elnakat Thomas H, Orr-Asman MA, Green LC, Boody RE, Matias K, Karve A, Hisada YM, Davis HW, Qi X, *et al*: mTOR kinase inhibition reduces tissue factor expression and growth of pancreatic neuroendocrine tumors. *J Thromb Haemost* 17: 169-182, 2019.
119. Conway JRW, Warren SC, Herrmann D, Murphy KJ, Cazet AS, Vennin C, Shearer RF, Killen MJ, Magenau A, Méléne P, *et al*: Intravital imaging to monitor therapeutic response in moving hypoxic regions resistant to PI3K pathway targeting in pancreatic cancer. *Cell Rep* 23: 3312-3326, 2018.
120. Sakamoto Y, Yamagishi S, Tanizawa Y, Tajimi M, Okusaka T and Ojima H: PI3K-mTOR pathway identified as a potential therapeutic target in biliary tract cancer using a newly established patient-derived cell panel assay. *Jpn J Clin Oncol* 48: 396-399, 2018.
121. Joechle K, Juma H, Thriene K, Hellerbrand C, Kulemann B, Fichtner-Feigl S, Lang SA and Guenzle J: Dual inhibition of mTORC1/2 reduces migration of cholangiocarcinoma cells by regulation of matrixmetalloproteinases. *Front Cell Dev Biol* 9: 785979, 2021.
122. Buzzoni R, Pusceddu S, Bajetta E De Braud F, Platania M, Iannacone C, Cantore M, Mambrini A, Bertolini A, Alabiso O, *et al*: Activity and safety of RAD001 (everolimus) in patients affected by biliary tract cancer progressing after prior chemotherapy: A phase II ITMO study. *Ann Oncol* 25: 1597-1603, 2014.
123. Ewald F, Grabinski N, Grottke A, Windhorst S, Nörz D, Carstensen L, Staufer K, Hofmann BT, Diehl F, David K, *et al*: Combined targeting of Akt and mTOR using MK-2206 and RAD001 is synergistic in the treatment of cholangiocarcinoma. *Int J Cancer* 133: 2065-2076, 2013.
124. Rodon J, Dienstmann R, Serra V and Tabernero J: Development of PI3K inhibitors: Lessons learned from early clinical trials. *Nat Rev Clin Oncol* 10: 143-153, 2013.
125. Stutfeld E, Aylett CH, Imseng S, Boehringer D, Scaiola A, Sauer E, Hall MN, Maier T and Ban N: Architecture of the human mTORC2 core complex. *Elife* 7: e33101, 2018.
126. Benavides-Serrato A, Lee J, Holmes B, Landon KA, Bashir T, Jung ME, Lichtenstein A and Gera J: Specific blockade of Rictor-mTOR association inhibits mTORC2 activity and is cytotoxic in glioblastoma. *PLoS One* 12: e0176599, 2017.
127. Werfel TA, Wang S, Jackson MA, Kavanaugh TE, Joly MM, Lee LH, Hicks DJ, Sanchez V, Ericsson PG, Kilchrist KV, *et al*: Selective mTORC2 inhibitor therapeutically blocks breast cancer cell growth and survival. *Cancer Res* 78: 1845-1858, 2018.
128. Waldner M, Fantus D, Solari M and Thomson AW: New perspectives on mTOR inhibitors (rapamycin, rapalogs and TORKinibs) in transplantation. *Br J Clin Pharmacol* 82: 1158-1170, 2016.
129. Yang C and Malarkannan S: Transcriptional regulation of NK cell development by mTOR complexes. *Front Cell Dev Biol* 8: 566090, 2020.
130. Yang W, Gorentla B, Zhong XP and Shin J: mTOR and its tight regulation for iNKT cell development and effector function. *Mol Immunol* 68: 536-545, 2015.
131. Singh Y, Garden OA, Lang F and Cobb BS: MicroRNA-15b/16 enhances the induction of regulatory T cells by regulating the expression of Rictor and mTOR. *J Immunol* 195: 5667-5677, 2015.
132. Moore KN, Hong DS, Patel MR, Pant S, Ulahannan SV, Jones S, Meric-Bernstam F, Wang JS, Aljumaily R, Hamilton EP, *et al*: A Phase Ib trial of prexasertib in combination with Standard-of-Care agents in advanced or metastatic cancer. *Target Oncol* 16: 569-589, 2021.
133. Zhu AX, Kudo M, Assenat E, Cattani S, Kang YK, Lim HY, Poon RT, Blanc JF, Vogel A, Chen CL, *et al*: Effect of everolimus on survival in advanced hepatocellular carcinoma after failure of sorafenib: The EVOLVE-1 randomized clinical trial. *JAMA* 312: 57-67, 2014.
134. Geissler EK, Schnitzbauer AA, Zülke C, Lamby PE, Proneth A, Duvoux C, Burra P, Jauch KW, Rentsch M, Ganten TM, *et al*: Sirolimus use in liver transplant recipients with hepatocellular carcinoma: A randomized, multicenter, open-label phase 3 trial. *Transplantation* 100: 116-125, 2016.
135. Chung V, Frankel P, Lim D, Yeon C, Leong L, Chao J, Ruel N, Luevanos E, Koehler S, Chung S, *et al*: Phase Ib trial of mFOLFOX6 and Everolimus (NSC-733504) in patients with metastatic gastroesophageal adenocarcinoma. *Oncology* 90: 307-312, 2016.
136. Joka M, Boeck S, Zech CJ, Seufferlein T, Wichert Gv, Licht T, Krause A, Jauch KW, Heinemann V and Bruns CJ: Combination of antiangiogenic therapy using the mTOR-inhibitor everolimus and low-dose chemotherapy for locally advanced and/or metastatic pancreatic cancer: A dose-finding study. *Anticancer Drugs* 25: 1095-1101, 2014.
137. Yu K, Toral-Barza L, Shi C, Zhang WG, Lucas J, Shor B, Kim J, Verheijen J, Curran K, Malwitz DJ, *et al*: Biochemical, cellular, and in vivo activity of novel ATP-competitive and selective inhibitors of the mammalian target of rapamycin. *Cancer Res* 69: 6232-6240, 2009.
138. Xiao Y, Liu P, Wei J, Zhang X, Guo J and Lin Y: Recent progress in targeted therapy for non-small cell lung cancer. *Front Pharmacol* 14: 1125547, 2023.



Copyright © 2024 Cao et al. This work is licensed under a Creative Commons Attribution-NonCommercial-NoDerivatives 4.0 International (CC BY-NC-ND 4.0) License.