

We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

6,900

Open access books available

186,000

International authors and editors

200M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.
For more information visit www.intechopen.com



Targeting the PI3K/AKT/mTOR Pathway in Cancer Cells

Isabella S. Guimarães, Nayara G. Tessarollo, Paulo C.M. Lyra-Júnior,
Diandra Z. dos Santos, Roger C. Zampier, Laura F.R.L. de Oliveira,
Krislayne V. Siqueira, Ian V. Silva and Leticia B.A. Rangel

Additional information is available at the end of the chapter

1. Introduction

The phosphoinositide 3-kinase (PI3K)/protein kinase B (AKT)/mammalian target of rapamycin (mTOR) pathway is a critical regulator of many essential physiological processes, but it also plays a key role in the malignant transformation of human tumors and their subsequent growth, metabolism, proliferation, and metastasis [1]. Previous studies have demonstrated that the PI3K/AKT/mTOR pathway is frequently activated in human cancers due to the somatic mutation and amplification of genes encoding key components [2,3]. In addition, aberrant PI3K/AKT/mTOR signaling activation also confers resistance to conventional therapies and is a poor prognostic factor for many types of cancers [4,5]. Several agents that target the PI3K/AKT/mTOR cascade elements are undergoing evaluation in preclinical and clinical studies. These include PI3K inhibitors, AKT inhibitors, mTOR catalytic site inhibitors, and dual PI3K-mTOR inhibitors. This chapter focuses on recent preclinical and clinical data on the efficacy of PI3K/AKT/mTOR pathway inhibitors either as monotherapy or in combination with conventional chemotherapy or others target drugs. Herein, we review four different classes of PI3K pathway inhibitors: PI3K inhibitors, AKT inhibitors, mTOR catalytic site inhibitors, and dual PI3K-mTOR inhibitors.

2. The PI3K/AKT/mTOR pathway

The PI3K/AKT/mTOR constitutes an important pathway downstream of growth factor tyrosine kinase receptors, thus regulating a plethora of biological processes as angiogenesis, proliferation, metabolism, survival, and differentiation [3]. Accumulating evidences indicate,

therefore, that alterations in the PI3K/AKT/mTOR axis play critical and multifaceted role in cancer pathogenesis and progression. Indeed, systematic analysis performed in 3.281 tumors from 12 cancer types of the Cancer Genome Atlas Pan-Cancer effort has revealed that elements of the PI3K/AKT/mTOR signaling pathway are among the highest frequently mutated genes in cancer, such as uterine corpus endometrioid, breast, colon, lung, head and neck, and ovarian carcinomas [4,5].

Subunit	Protein	Gene name (human)
Class I		
Class IA		
Catalytic	p110 α	PIK3CA
	p110 β	PIK3CB
	p110 δ	PIK3CD
Regulatory	p50 α , p55 α , p85 α	PIK3R1
	p85 β	PIK3R2
	p55 γ	PIK3R3
Class IB		
Catalytic	p110 γ	PIK3CG
Regulatory	p101	PIK3R5
	p84, p87	PIK3R6
Class II		
Catalytic	PI3KC2 α	PIK3C2A
	PI3KC2 β	PIK3C2B
	PI3KC2 γ	PIK3C2G
Class III		
Catalytic	Vps34	PIK3C3
Regulatory	Vps15	PIK3R4

Table 1. The PI3K proteins family

PI3K is a heterodimer of its catalytic and regulatory subunits and has been classified as class I, II, and III. Class I PI3K is constituted by four 110-kDa catalytic subunits and two main regulatory domains, which is subdivide in class IA and IB. Class IA PI3K (PI3K α , β , and δ) is activated by receptors with tyrosine kinase activity, and class IB PI3K (PI3K γ) is activated by G protein-coupled receptors. The class IA enzymes are dimers of p110 α , p110 β , or p110 δ catalytic subunits and the regulatory subunits p85 α (or its splice variants p55a and p50a), p85 β , p55 γ , p101, or p84 [6,7]. In turn, class IB enzymes are dimers of p110 γ catalytic subunit and either p101 or p84 (also known as p87PIKAP) regulatory subunits [8]. The four class I

catalytic isoforms share overlapping but distinct functions. Although the expression of p110c and p110d isoforms seems to be confined to immune cells, p110a and p110b are ubiquitously expressed but exhibit isoform-specific cell-type- and context-dependent requirements, thus being involved in a wide range of cellular effects [9–13]. Class II PI3K (PI3KC2) subfamily has additional domains in both N- and C-terminal extensions and exists as 3 isoforms, PI3K-C2 α , PI3K-C2 β , and PI3K-C γ [14]. On the other hand, class III PI3K occurs as a single isoform constituted by the catalytic subunit Vps34p and regulatory subunit Vps15 [14] (Table 1).

The PI3K family recruits effector proteins, altering their localization, activity, and conformation. There are some binding proteins domains that mediate such events [14]. The best-characterized domains among them are FYVE (Fab 1, YOTB, Vac 1, EEA1) [15–17], PH (pleckstrin homology) [18], and PX (Phox) [19–23]. Nonetheless, the peculiar composition of the three PI3K subfamilies results in the activation of distinct cellular functions.

In brief, after activation by receptor tyrosine kinases, including members of platelet-derived growth factor receptor, the insulin and insulin-like growth factor 1 (IGF-1) receptors and human epidermal growth factor receptor family (EGFR and HER2), PI3K phosphorylates phosphatidylinositol 4,5-trisphosphate (PIP₂) to generate phosphatidylinositol 3,4,5-trisphosphate (PIP₃) [24]. In physiological conditions, the level of PIP₃ is strictly regulated by PTEN (phosphatase and tensin homolog), a phosphatase that specifically catalyzes the dephosphorylation of PIP₃, converting PIP₃ back to PIP₂, thus constituting an important endogenous-negative feedback loop of the PI3K signaling pathway [25,26]. The lipid product of PI3K, PIP₃, recruits a subset of signaling proteins with PH domains to the membrane, including 3-phosphoinositide-dependent protein kinase (PDK1) and AKT, resulting in its phosphorylation at threonine-308 and activation [24].

In both physiological and pathological conditions, AKT exists in three isoforms in mammals: AKT1, AKT 2, and AKT 3 [27,28]. AKT phosphorylates tuberous sclerosis complex 2 (TSC2), thereby inhibiting the GTPase activity of the TSC1/TSC2 complex and enabling mTOR activation by RAS homologue enriched in brain (RHEB), thus allowing signal propagation [26,29]. mTOR exists in two different structural protein complex: mTORC1 and mTORC2, each of which is expressed in different subcellular compartments, therefore affecting their activation and function. mTORC1 complex is composed of a catalytic subunit mTOR, regulatory-associated protein of mTOR (RAPTOR), mammalian lethal with SEC13 protein 8 (MLST8), and the noncore components PRAS40 and DEP domain-containing mTOR-interacting protein (DEPTOR). Once activated, mTORC1 leads to increased protein synthesis via its effectors, named translation-regulating factors ribosomal S6 kinase-1 (S6K-1) and eukaryote translation initiation factor 4E binding protein-1 (4EBP-1). S6K-1 and 4EBP1 are major regulators of protein translation [30]. On the other hand, mTORC2 is composed by rapamycin-insensitive companion of mTOR (RICTOR), MLST8, and mammalian stress-activated protein kinase interacting protein 1 (SIN1). The function of mTORC2 remains not fully understood, but it is required to phosphorylate AKT at serine-473, thus resulting in its maximal activation [31]. Of clinical relevance, differently from mTORC1, mTORC2 is insensitive to rapamycin inhibition, opening an avenue for drug discovery in face of the development of resistance by cancer cells against first-generation mTOR inhibitors (rapalogs) that particularly target mTORC1 [32] (Figure 1).

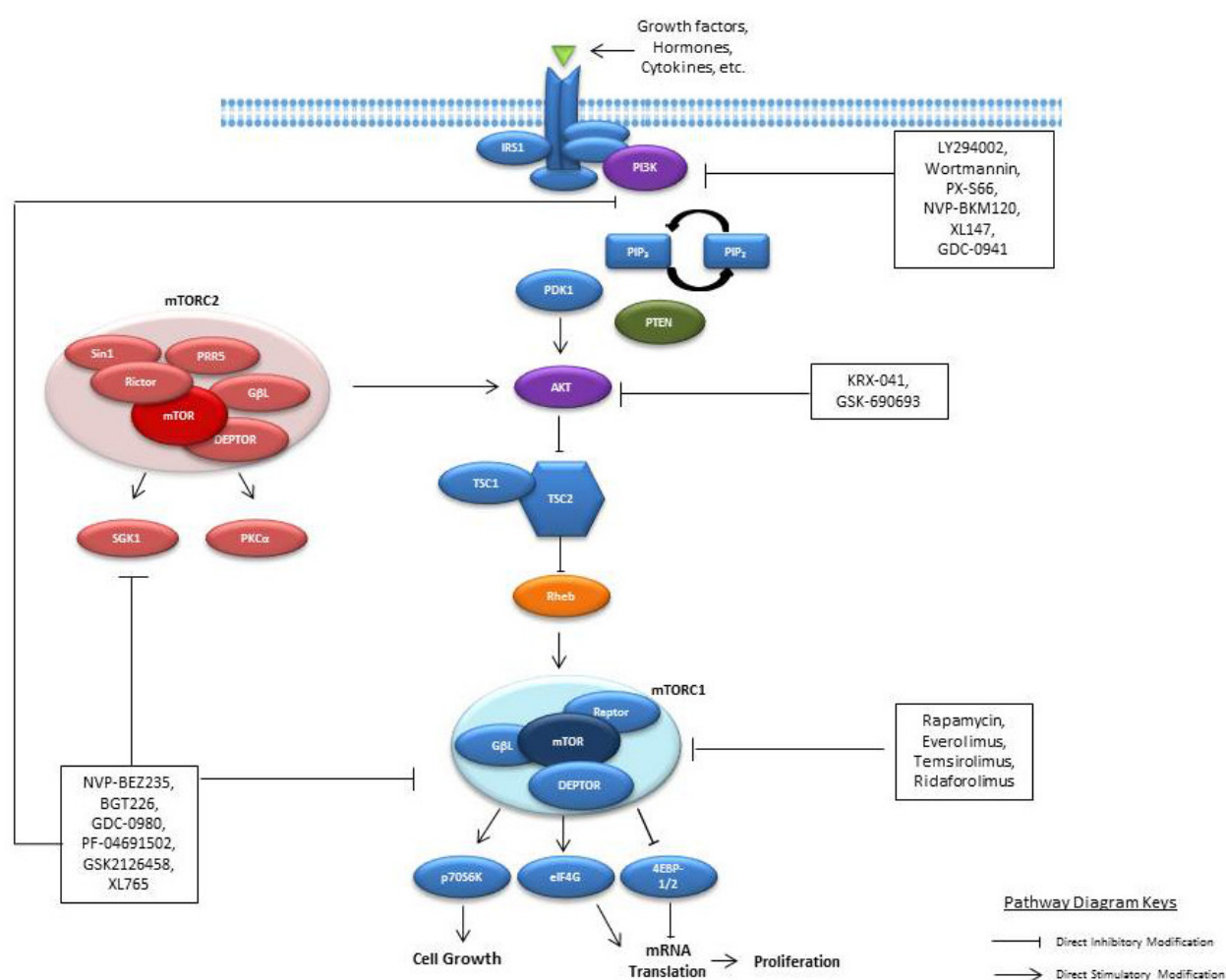


Figure 1. Overview of PI3K/AKT/mTOR signaling pathway and some inhibitors of this pathway in clinical studies. The activation of the PI3K by receptor tyrosine kinases promotes conversion of PIP₂ to PIP₃. PTEN dephosphorylates PIP₃, negatively regulating the PI3K signaling. The phosphorylation and activation of AKT impacts many downstream effectors, such as mTORC1, and finally leads to multiple cellular processes.

3. The role of PI3K/AKT/mTOR in cancer

Somatic mutations and/or gains and losses of genes are possible genetic alterations affecting the PI3K/AKT/mTOR pathway in different solid and hematological tumors [33,34]. Indeed, PI3K pathway can be activated by direct upstream signs and can be intrinsically activated due to gain of functional mutations or amplifications in PIK3CA (p110 subunit), mutations in PIK3R (p85 subunit), and mutations or amplifications in one of the AKT isoforms or loss of PTEN [35]. Loss of PTEN via inactivating mutations, due to either copy number loss or homozygous deletions, is associated with both resistance to chemotherapy and reduced survival of human patients [3].

PIK3CA mutations in primary breast tumors have been associated with lymph node metastases and overexpression of ER, PR, and HER2 [36]. Furthermore, the presence of activating

PI3KCA mutations and loss of PTEN in HER2-overexpressing cancers is correlated with a lower response to trastuzumab and lapatinib [37]. In non-small cell lung cancer, the downregulation of PTEN is also related with poor prognosis [38,39]. In ovarian cancer, PI3K/AKT/mTOR molecular alteration appears to be histological subtype specific. Studies have described amplifications in PIK3CA, amplifications of one of the AKT isoforms, and PTEN deletions in 20%, 15%, and 5% of the high grade serous ovarian cancer (HGSOC) cases, respectively [40,41]. The individual mutations, rare events in HGSOC, are prevalent in low grade serous, mucinous, endometrioid, and clear cell ovarian cancer; 20% of endometrioid and 35% of clear cell ovarian tumors display these PIK3CA mutations [42,43]. Besides, copy number changes in the genes encoding PIK3CA and PIK3CB subunits have been associated with a poor prognosis, and the inhibition of PI3K/mTOR was found to delay tumor growth and prolong survival [44,45].

Moreover, mutations of mTOR itself and/or in components of mTOR-related signaling pathways have frequently been described in human malignant diseases [46-48]. Different genetic lesions that mediate mTORC1 activation have diverse consequences: PTEN loss uncouples mTORC1 activation from growth factor signaling; liver kinase B1/serine/threonine kinase 11 (LKB1/STK11) mutations allow mTORC1 activation despite nutrient deprivation in poorly vascularized tumors; P53 mutations uncouple DNA damage from the inhibition of bioenergetic processes and cell cycle arrest [49]; and hyperactivation of S6K-1, 4EBP1 and eIF4E, and cancer growth by activating the lipid and protein biosynthesis. Furthermore, the increased phosphorylation of mTOR is associated with acquired cisplatin resistance, and AKT signaling has been implicated in primary platinum resistance [50]. In fact, AKT or mTOR inhibitors likely restore chemosensitivity to platinum derivatives *in vitro* and in xenograft models [51,52].

These molecular alterations, in addition to the druggability of the components of the PI3K/AKT/mTOR signaling cascade, suggest that targeting the pathway might represent a useful treatment strategy in the fight against cancer.

4. PI3K inhibitors in cancer therapy

As aforementioned, PI3K/AKT/mTOR pathway has been implicated in tumorigenesis, promotion of cell survival, angiogenesis, cellular invasion, tumor growth, and the acquisition of chemoresistant phenotype by cancer cells [1]. Currently, more than fifty PI3K/AKT/mTOR axis inhibitors are in different stages of development, with a great number of such inhibitors reaching clinical trials [53]. Analogs of rapamycin (inhibitors of mTORC1), temsirolimus and everolimus, are currently in the lead, having already been approved by the Food and Drug Administration (FDA) as anticancer agents [54-56]. The PI3K/AKT/mTOR pathway inhibitors are summarized in Table 2.

4.1. PI3K inhibitors

PI3K inhibitors can be divided in isoform-specific inhibitors or pan-PI3K inhibitors. pan-PI3K inhibitors target all class IA PI3Ks in tumor cells, whereas isoform-specific inhibitors were

developed to decrease toxicity and might be particularly effective in cancers with PIK3CA mutations, for example.

The first-generation of PI3K inhibitors include wortmannin, a fungal metabolite isolated from *Penicillium wortmannin* that irreversibly inhibits p110 by reacting covalently with the catalytic site [57], and LY294002, a synthetic, competitive, and reversible inhibitor of the ATP binding site of PI3K [58]. Both agents achieve significant antiproliferative and pro-apoptotic effects in preclinical *in vitro* and *in vivo* studies. However, unfavorable pharmacokinetic properties, insolubility in water, high levels of toxicity, and lack of selectivity for oncogenic isoforms of Class I PI3K limit its use in clinical trials [59,60]. Although this limiting features for their clinical use, wortmannin and LY294002 have served as important research tool for elucidating diverse signal transduction processes involving PI3K pathway and has spawned a new generation of PI3K inhibitors [61] (Table 2).

Currently, water-soluble wortmannin conjugates are being developed to overcome this issue. PX-866 is a semisynthetic analog of wortmannin with potent, irreversible, pan-class I PI3K inhibitory property against p110- α , p110- δ , and p110- γ enzymes in biochemical assays [62]. In preclinical studies, the compound alone or in combination with chemotherapy (cisplatin), radiotherapy, and targeted cancer drugs (gefitinib) exhibited *in vivo* antitumor activity against numerous mouse xenograft models of human cancers [62,63]. In addition, a phase I study in eighty-four patients with advanced solid tumors showed that PX-866 is well tolerated. The most frequent study drug-related adverse events were gastrointestinal disorders, with diarrhea being the most common [64]. PX-866 is being currently tested in a combination phase I/II studies with cetuximab (NCT01252628) in squamous cell carcinoma of the head and neck (SCCHN) and in metastatic colorectal carcinoma. Furthermore, more two phase I/II studies with PX866 are ongoing: with docetaxel (NCT01204099) in non-small cell lung cancer and SCCHN and in combination with vemurafenib in patients with advanced melanoma (NCT01616199).

Buparlisib (NVP-BKM120) is an oral highly specific pan-class I PI3K inhibitor with inhibitory property against p110- α , p110- β , p110- δ , and p110- γ enzymes [65]. The compound is also active against activating p110 α somatic mutations but does not significantly inhibit the related class III and class IV PI3K kinases. In preclinical cancer studies, buparlisib has shown antiproliferative and proapoptotic activity against a panel of 353 cell lines that display different genetic abnormalities that promote PI3K pathway activation [66]. *In vivo* studies have also shown that buparlisib potently inhibits the growth of human xenografts models and behaves synergistically when combined with cytotoxic agents such as temozolomide, alkylating agent, and docetaxel, antimetabolic drug, or with targeted agents such as HER2 and mitogen-activated protein kinase kinase (MEK) inhibitors [66].

A phase I dose-escalation study in thirty-five patients with advanced-stage solid tumors showed that buparlisib is a safe and well-tolerated drug with favorable pharmacokinetic properties. The major treatment-related adverse events included rash, hyperglycemia, diarrhea, anorexia, mood alteration, nausea, fatigue, pruritus, and mucositis [67]. Importantly, hyperglycemia was more common at higher doses and represents a class effect of the inhibition of PI3K signaling, commonly observed with other PI3K/AKT/mTOR pathway inhibitors [67].

Later, phase I dose-escalation and expansion study of buparlisib was performed in eighty-three patients with advanced solid tumors demonstrating that buparlisib was well tolerated up to 100 mg/day and showed preliminary activity in patients with advanced cancers [68]. This subsequently led to the initiation of several clinical trials in multiple cancer types, such as non-small cell lung cancer, prostate cancer, breast cancer, colon cancer, and glioblastoma multiform (GBM).

BASALT-1, an ongoing phase II trial (NCT01297491), is investigating the efficacy of single-agent buparlisib in patients with metastatic non-small cell lung cancer with PI3K pathway activation. Furthermore, phase Ib/II is under evaluation in patients with advanced non-small cell lung cancer of different histotype, testing buparlisib in combination with other targeted agents such as everolimus (NCT01470209), erlotinib (NCT01487265), MEK inhibitor (NCT01363232), or in combination with standard chemotherapeutic drugs, such as docetaxel (NCT01911325), gemcitabine, and cisplatin (NCT01971489) and carboplatin and paclitaxel (NCT01820325).

At present, several active, not recruiting, and recruiting clinical trials are being conducted in all the biological subsets of breast cancer, including combinations with endocrine therapy, anti-HER2 agents, poly (ADP-ribose) polymerase (PARP) inhibitors, and chemotherapy with buparlisib. Two large phase III studies (BELLE-2 and BELLE-3) (NCT01610284, NCT01633060) are investigating the combination of buparlisib plus fulvestrant in postmenopausal women with hormone receptor-positive/HER2-negative breast cancer after failure of aromatase inhibitor alone or aromatase inhibitor plus mTOR inhibitor treatment, respectively. Another ongoing clinical study is BELLE-4, a placebo-controlled phase II trial of buparlisib with paclitaxel in the first-line treatment of HER2-negative metastatic breast cancer (NCT01572727). Buparlisib has also been evaluated in a phase II study of paclitaxel plus trastuzumab in HER2-overexpressing breast cancer (NCT01816594).

Pilaralisib (XL147) is an oral pan-class I PI3K inhibitor (α , β , γ , and δ) through reversible, competitive inhibition with ATP for p110- α , - δ , - γ , and - β enzymes [69]. *In vitro* tests revealed that pilaralisib inhibits the formation of PIP₃ in the membrane and phosphorylation of AKT and S6K-1 in multiple tumor cell lines with diverse genetic alterations in PI3K pathway [70]. Moreover, in mouse xenograft models, oral administration of pilaralisib results in significant tumor growth inhibition and combination with chemotherapeutic agents improved the growth-inhibitory effect observed with the single agents [71]. Based on this preclinical rationale, pilaralisib has been evaluated in phase I/II clinical trials.

In a phase I dose-escalation trial of sixty-nine patients with advanced solid tumors, pilaralisib was tolerable at doses associated with PI3K pathway inhibition, and the most frequent drug-related adverse events included dermatologic toxicities, diarrhea, nausea, and decreased appetite [72]. However, a phase I dose-escalation study of pilaralisib with erlotinib in patients with solid tumors showed that combination had limited antitumor activity with moderate inhibition of PI3K, MAPK and EGFR pathways [73]. Moreover, phase I/II study of pilaralisib in combination with trastuzumab or trastuzumab plus paclitaxel in trastuzumab-refractory HER2-positive metastatic breast cancer related that no responses were observed in patients treated with pilaralisib plus trastuzumab while clinical activity was observed in paclitaxel arm

[74]. Additional clinical evaluation of this PI3K inhibitor is ongoing in phase I/II studies (NCT01587040).

Pictilisib (GDC-0941) is another potent, selective, and orally bioavailable inhibitor of pan-class I PI3K. In biochemical assays, pictilisib demonstrates selectivity over a large panel of protein kinases and PI3K family kinases, including mTOR and DNA-dependent protein kinase (DNA-PK) [75]. Interestingly, pictilisib induces apoptosis in a subset of human tumor cell lines and potently inhibited tumor growth in xenograft models, including those with mutations in PI3K, PTEN, and K-Ras [76]. Significant *in vivo* antitumor activity has also been observed when administered orally in combination with other anticancer drugs, for example, docetaxel and MEK inhibitor U0126 [77-80].

In a first-in-human phase I study of pictilisib in sixty patients with advanced solid tumors, the most frequently reported drug-related adverse events were nausea, fatigue, and rash [81]. Importantly, one patient with V600E BRAF-mutant melanoma and another with platinum-refractory ovarian cancer exhibiting PTEN loss and PIK3CA amplification demonstrated partial response [81]. Pictilisib is currently under evaluation in several phase I/II clinical trials, mainly in non-small cell lung cancer and breast cancer (NCT01918306, NCT01740336, NCT01493843, and NCT00974584).

One strategy to achieve significant pathway inhibition clinically with tolerable adverse effect profile is the use of isoform-specific PI3K inhibitors. As aforementioned, each isoform has distinct role in normal physiological processes and disease (Table 1). PI3K catalytic subunit p110 α is predominantly responsible for mediating growth factor signaling from receptor tyrosine kinases and is a frequent genetic driver (*PIK3CA* mutations) in several cancers [82]. However, p110 α is dispensable for PI3K pathway activation in tumors lacking PTEN. Thus, these cells depend largely on p110 β to activate the pathway [82,83]. Preclinical tests showed that p110 β -selective inhibitors had a significantly greater activity in cell lines with PTEN null than in those with PTEN intact, although, some PTEN-intact cell lines were sensitive and a number of cell lines lacking PTEN were resistant [84]. GSK-2636771 is a PI3K p110 β -selective inhibitor currently in phase I studies in subjects with advanced solid tumors with PTEN deficiency (NCT01458067). Moreover, PI3K δ is predominantly expressed in leukocytes and control immune responses [85]. Idelalisib (CAL-101), a highly specific PI3K δ inhibitor, was the first isoform-specific PI3K inhibitors approved for cancer treatment [86].

Alpelisib (NVP-BYL719) is an oral inhibitor that selectively targets PI3K p110 α equipotent against the wild type and the most common somatic mutations of p110 α [87]. NVP-BYL719 has been the first PI3K α -selective inhibitor to enter in clinical trials after positive preclinical investigations. *In vivo* studies have demonstrated dose-dependent antitumor activity of NVP-BYL719 in PIK3CA-mutant or PIK3CA-amplified tumor xenograft models, such as ovarian, breast, and head and neck cancers [88, 89]. Preliminary results of phase I study performed in patients with advanced solid tumors carrying *PIK3CA* gene alterations demonstrated that NVP-BYL719 has a favorable safety profile with manageable toxicities, as hyperglycemia, nausea, diarrhea, decreased appetite, vomiting, and fatigue [90]. To date, more than fifteen clinical trial is ongoing in order to evaluate the combination of NVP-BYL719 with several agents, such conventional cytotoxic drugs (paclitaxel, cisplatin, and irinotecan) and target

drugs (cetuximab, olaparib, and trastuzumab) in a subset of cancers (NCT02051751, NCT01822613, NCT01602315, NCT01623349, and NCT02167854)

Taselisib (GDC-0032) is a PI3K inhibitor with higher affinity for mutated PI3K α with reduced inhibitory activity against PI3K β [91]. Preclinical studies show that taselisib has enhanced activity against PI3K α isoform mutant cancer cell lines [92]. In an ongoing phase I study, taselisib has been well tolerated with hyperglycemia and fatigue being the dose-limiting toxicities [93]. This selectivity profile and excellent pharmacokinetic properties allowed fewer clinical studies with GDC-0032. Currently, several clinical studies are ongoing to evaluate the combination of taselisib with endocrine therapy, trastuzumab, and conventional chemotherapy in breast cancer (NCT02285179, NCT02390427, and NCT01862081). In addition, a phase I study is currently ongoing in taselisib with CDK4/6 inhibitor, palbociclib, in advanced solid tumors and breast cancer (NCT02389842).

Idelalisib was approved in 2014 in the United States and European Union for the treatment of three indolent B-cell neoplasms: relapsed chronic lymphocytic leukemia, in combination with rituximab, relapsed follicular B-cell non-Hodgkin's lymphoma, and relapsed small lymphocytic lymphoma (as monotherapy) [94]. In lymphoid cell lines and primary patient samples, idelalisib abrogates PI3K/AKT/mTOR signaling and promotes apoptosis [95,96]. The first phase I trial in healthy volunteers established the bioavailability and safety of idelalisib [97]. Another phase I study in patients with relapsed/refractory mantle cell lymphoma reported the most common adverse events, which includes diarrhea, nausea, pyrexia, fatigue, rash, upper respiratory infection, pneumonia and alanine transaminase, or aspartate transaminase elevations [98]. To date, about twenty-five clinical trials are ongoing with idelalisib. A phase I/II trial studies aimed evaluated idelalisib in combination with lenalidomide and rituximab in patients with relapsed or refractory mantle cell lymphoma (NCT01838434). In addition, idelalisib is being evaluated in combination with rituximab in adults with previously treated indolent non-Hodgkin lymphoma (NCT01732913).

5. AKT inhibitors in cancer therapy

AKT inhibitors constitute another class of drugs that has gained recent interest. As discussed previously, AKT is involved in the regulation of various signaling downstream pathways involved in cell survival, growth, proliferation, metabolism, and angiogenesis. AKT inhibition promotes decreasing cancer cell survival by preventing signal transduction through its downstream effectors. In addition, targeting AKT is an interesting pharmacological approach due to the AKT activation in consequence of the feedback loop release when mTOR is inhibited.

AKT inhibitors can be grouped into three classes, including lipid-based phosphatidylinositol (PI) analogs, ATP-competitive inhibitors (catalytic inhibitors), and allosteric inhibitors. To date, the most developed inhibitor of AKT is perifosine (KRX-0401), a lipid-based inhibitor. Perifosine is an allosteric inhibitor that targets the PH domain of AKT, thereby preventing its translocation to the plasma membrane required for pathway activation [99]. Perifosine has demonstrated great efficacy *in vitro* and *in vivo* against several human cancers such as breast,

osteosarcoma, ovarian, multiple myeloma, leukemia, and glioma [100,101]. Additional *in vitro* data demonstrate synergistic effects of perifosine and traditional chemotherapeutic agents such as paclitaxel and cisplatin in ovarian cancer [102,103], etoposide in leukemia cells [104], doxorubicin in multiple myeloma cells [105], and gemcitabine in pancreatic cells [106].

Despite these encouraging preclinical studies, results from phase I/II clinical trials of perifosine as single agent in a various tumor types (metastatic breast cancer, metastatic head and neck cancer, locally advanced soft tissue sarcoma, prostate cancer, and metastatic

In behalf of the poor efficacy of perifosine as a single agent observed in most tumor types evaluated thus far, efforts have been made to combine this drug with target agents and chemotherapy. Phase I studies have now confirmed the safety of these combinations with different agents, including sorafenib in patients with Hodgkin lymphoma and taxanes in high-grade epithelial ovarian cancer [112,113]. Currently, one clinical trial with perifosine is recruiting patients, a phase II study with perifosine and temsirolimus in patients with malignant gliomas (NCT02238496).

GSK-690693 is a potent ATP-competitive AKT inhibitor selective for all three AKT isoforms versus the majority of kinases assessed by biochemical tests [114]. GSK690693 displayed antiproliferative activity *in vitro* and *in vivo* models of ovarian, breast, and prostate cancer [114]. The compound has entered phase I trials for refractory hematologic malignancies but was withdrawn prior to enrolment (NCT00666081).

6. mTOR inhibitors in cancer therapy

6.1. Rapamycin and its derivatives

As discussed previously, mTOR is involved in many cell signaling pathways, and clinical trials for cancer treatment showed that tumor cells with mutations in p53 or PTEN are susceptible to mTOR inhibitors [115]. mTOR inhibitors are categorized in first- and second-generation presenting a wide variety of target and mechanism. The first-generation mTOR inhibitors include rapamycin and its analogs that employ allosteric mechanism to block, whereas the second-generation mTOR inhibitors (AZD8055, Torin1, PP242, and PP30) have as target ATP binding site to impede kinase activity of both mTORC1 and mTORC2 [116].

Rapamycin, discovered in 1975, is a macrocyclic lactone isolated from the soil bacterium *Streptomyces hygroscopicus*, and it has clinical applications including antifungal, immunosuppressant, and anticancer proprieties [117,118]. FDA approved this drug in 1997 for prevention of host-rejection during kidney transplants [119]. Preclinical studies have shown that rapamycin presents strong antiangiogenic and antiproliferative properties against a variety of human cancers such as the phase II study, which showed rapamycin potentiates the effect of paclitaxel in endometrial cancer cell lines [120].

Three different mechanisms of action have been proposed: first, the binding of the FKBP-12–rapamycin complex to mTOR that could lead dephosphorylation of downstream effector

molecules such as S6K-1 and 4EBP1 [121]; second, the FKBP-12–rapamycin complex competes with phosphatidic acid to bind to the FRB domain of mTOR, blocking mTOR kinase function [122]; and third, the FKBP-12–rapamycin complex binds to mTOR and destabilizes the mTOR–raptor–4EBP1/S6K-1 scaffold complex, leading to dephosphorylation of S6K-1 and 4EBP1 [123,124].

This inhibitor has limited bioavailability due to its poor aqueous solubility. In an effort to improve its pharmacokinetics, several rapamycin analogs, named rapalogs, have been developed, such as temsirolimus (CCI-779), everolimus (RAD001), and ridaforolimus (MK-8669/AP23573) [125-127].

Some studies have shown that these compounds are able to disrupt the mTORC2 complex in a dose-, time-, and cell type-dependent manner [24,128,129]. A possible mechanism by which rapamycin and rapalogs could inhibit mTORC2 relies on the interaction of newly synthesized mTOR molecules and rapamycin/rapalogs-FKBP12 complexes. In turn, this interaction would prevent mTOR from the interaction with RICTOR, thus inhibiting mTORC2. Indeed, it has been shown that prolonged exposure of cancer cells to rapamycin can promote its binding to mTOR before the assembly of the mTORC2 complex, with subsequent inhibition of the AKT-mediated signaling [24].

Rapamycin and its derivatives exhibit a safe toxicity profile, being the side effects of skin rashes and mucositis dose dependent [130]. Other symptoms commonly described are fatigue, nausea, anemia, hypertriglyceridemia, hypercholesterolemia, and neutropenia [131]. Furthermore, temsirolimus and sirolimus are associated with significant rate of pulmonary toxicity [130,131]. Rare side effects of the aforesaid drugs include interstitial lung disease, risk of secondary lymphoma, and reactivation of latent infections [35].

Everolimus (Afinitor®), the oral mTOR inhibitor, has been approved by the FDA in 2009 for advanced renal cell cancer. Everolimus exhibit strong antiangiogenic and antiproliferative activity against various human cancer such as metastatic or unresectable pancreatic neuroendocrine tumors, subependymal giant cell astrocytoma [132], metastatic renal cell carcinoma, and advanced estrogen receptor (ER)-positive [133] and human epidermal growth factor receptor-2 (HER2)-negative breast cancer [134].

Several studies have been conducted to analyze the effectiveness of rapamycin and rapalogs alone and in combination with standard chemotherapy, hormonal therapy such as anti-VEGF inhibitors in the treatment of several types of cancers such breast, ovarian, cervical, and endometrial. Phase II studies are ongoing in order to test everolimus in combination with chemotherapy (cisplatin and gemcitabine) in patients with metastatic triple negative breast cancer (NCT01939418 and NCT01931163). In addition, a recent study of breast cancer (BOLERO-3) demonstrated that the combination of everolimus with trastuzumab and vinorelbine significantly prolonged progression-free survival (PFS) in patients with trastuzumab-refractory and taxane-pretreated, HER2-positive advanced breast cancer [135]. Moreover, another breast cancer study, BOLERO-1, evaluated patients treated with paclitaxel and trastuzumab with or without everolimus as first-line therapy [136]. Furthermore, clinical studies have

evaluated the aromatase inhibitor letrozole in combination with everolimus in patients with metastatic endometrial carcinoma (NCT01068249) and breast cancer (NCT00107016).

Temsirolimus (Torisel®), the first rapamycin analog to be FDA approved as an anticancer drug, is an intravenous injection drug and gets converted into rapamycin *in vivo* [137]. This drug was valued with bevacizumab or in combination with chemotherapeutic agents in endometrial cancer cell lines, and results showed the increase progesterone mRNA expression and inhibition of ER mRNA expression [138,139]. Also, preliminary phase II study using temsirolimus in patients with metastatic cervical cancer showed positives results [140]. Another phase II clinical study (NCT01196429) evaluates additional effects of the temsirolimus combined with paclitaxel/carboplatin therapy have been conducted in patients with stages III/IV clear cell adenocarcinoma [141]. However, some studies failed to show the efficiency of temsirolimus in patients with persistent/recurrent epithelial ovarian cancer/primary peritoneal cancer showing a modest activity of this mTOR inhibitor, and the results were insufficient to justify further study in a phase III [142].

Ridaforolimus (MK-8669/AP23573), a non-rapamycin prodrug, is available in both oral and intravenous formulations. This mTOR inhibitor is actively being evaluated as either monotherapy or in combination with other therapies for treatment of various cancers, including sarcomas, endometrial, prostate, breast, and non-small cell lung cancer [143]. Studies had been conducted in patients with advanced endometrial cancer and clinical benefit response was reported in 33% of the patients [144]. Another phase II study using oral ridaforolimus in patients with advanced or recurrent endometrial cancer also showed partial response in 7.7% patients [145].

Although clinically promising, the efficacy of rapalogs is partially limited by the negative feedback loops in the mTOR pathway. With this regard, the exclusive inhibition of the mTORC1 complex by the rapalogs compromises the S6K-1-mediated feedback loop towards IRS-1, resulting in the activation of both the PI3K/AKT and the mitogen-activated protein kinase/extracellular signal-regulated kinases (MAPK/ERK) pathways, hence promoting compensatory cell survival, and the acquisition of chemoresistant phenotype [127,146,147]. Efforts have been made to overcome the previously mentioned clinical limitation by means of developing new generation mTOR inhibitors, which inhibit the catalytic activity of both mTORC1 and mTORC2 complexes.

7. ATP-competitive inhibitors

Although rapamycin is a potent allosteric mTORC1 inhibitor with clinical applications, a second-generation ATP-competitive inhibitor have been developed, including Torin1, Torin2, PP242, PP30, KU0063794, WAY-600, WYE-687, WYE-354, XL-388, INK-128, AZD-2014, AZD8055, and OSI-027 [148-153]. The ATP-competitive inhibitors of mTOR directly inhibit the mTOR kinase activity, affecting both mTORC1 and mTORC2 complexes simultaneously and suppress AKT activity.

ATP-competitive mTOR inhibitors represent a promising new approach to target the pathway with potentially greater tolerability and efficacy than rapamycin. It has been shown that ATP inhibitors displayed dramatic antiproliferative activity across a range of cancer cell lines [151,154,155].

Studies have been conducted with PP242 in colon cancer cells *in vitro* and *in vivo* showed decrease cell growth alone or in combination with MEK inhibitors [156]. Another ATP competitive inhibitor, Torin2, was developed to overcome the pharmacological limitations of Torin1 and it is a potent inhibitor of ATR, ATM, and DNA-PK [157,158]. Lung cancer cell treatment with Torin2 resulted in a prolonged block in negative feedback and consequent threonine-308 phosphorylation on AKT. These effects were associated with strong growth inhibition *in vitro* [159].

Studies conducted by Rodrik-Outmezguine and colleagues [160], comparing mTORC1 inhibition with rapamycin and AZD8055, revealed that rapamycin treatment led to an almost complete loss in the mTORC1 phosphorylation of S6K-1 (threonine-389) and increased phospho-AKT (serine-473). In contrast, AZD8055 treatment led to reductions in phospho-S6K-1 (threonine-389), phospho-4EBP1 (threonine-37/40, threonine-65, and threonine-70), and phospho-AKT (serine-473). Thereby, AZD8055 was a better inhibitor of mTORC1 in comparison to rapamycin. *In vivo* studies indicated that AZD8055 can inhibit tumor growth and AZD8055 showed promise as a therapeutic agent.

At present, there are several clinical trials focused on the examination of new agents, such as AZD-8055 (NCT00731263), OSI-027 (NCT00698243), and INK128 (NCT02142803), in a variety of human hematological malignancies and solid tumors, including breast cancer. Also some studies were conducted using GSK795 in patients with advanced platinum-resistant ovarian and showed interesting results as tumor regressions and CA125 decreases [161]. Phase I study are ongoing to evaluate the safety and toxicity profile of AZD2014 in combination with paclitaxel in patients with ovarian cancer (NCT02193633).

Despite the clinical improvements observed with the ATP-competitive inhibitor when compared to the rapalogs, the literature still acknowledges significant limitations that outcome from compensatory cellular events. With this regard, it has been found that loss of the feedback on PI3K results in compensatory activation of the MAPK/ERK cascade by mTOR downstream effectors, such as 4EBP1/eIF4E, maintaining cell proliferation [162]. Furthermore, it has been shown that chronic inhibition of mTORC2 induces the activation of AKT by its phosphorylation mediated by PDK-1, even in the absence of the priming serine-473 phosphorylation. Altogether, the referred mechanisms ultimately drive the acquisition of the resistant phenotype by the cancer cells [154,163].

8. Dual mTOR/PI3K inhibitors

Scientists have explored to shed light on strategies to overcome the limitations by concomitantly targeting two molecules in the PI3K/AKT/mTOR pathway, PI3K and mTOR, whereas the

resistance mTOR inhibitors could arise via feedback PI3K activation. This molecular knowledge has stimulated the development of new inhibitors termed dual PI3K-mTOR inhibitors that include NVP-BEZ235, XL765, BGT226, PI-103, PF-04691502, PKI-587, and GDC-0980 [164-170]. Comparing with the other types of PI3K pathway inhibitors, dual PI3K-mTOR inhibitors have the possible advantage of inhibiting all PI3K catalytic isoforms, mTORC1 and mTORC2 [171]. Therefore, these inhibitors may effectively turn off this pathway completely and display best efficacy in feedback inhibition normally observed with mTORC1 inhibitors [172]. However, it is not clear that dual PI3K-mTOR inhibitors will be tolerable at doses that effectively inhibit all p110 isoforms and mTOR [171].

The potential clinical value of the dual PI3K/mTOR inhibitors has been demonstrated by their significant inhibition of cell growth, the induction of apoptosis and/or autophagy [173] in a variety of tumor cancer cells [174-176]. In addition, these inhibitors have shown powerful effects in xenograft models of breast cancer [177], pancreatic cancer [178], melanoma [179], multiple myeloma [180], and RCC [181].

In agreement, dual PI3K/mTOR inhibitors have entered clinical trials either monotherapy or polytherapy. A single agent includes BEZ235/NVP-BEZ235 (NCT00620594) and BGT226 (NCT00600275 and NCT00742105) in advanced solid tumors and breast cancer, GDC-0980 (NCT00854126, NCT00854152, and NCT01455493) in non-Hodgkin lymphoma and endometrial carcinoma, and PF-04691502 (NCT00927823) and GSK2126458 (NCT00972686 and NCT01248858) in solid tumors. In combination with other agents, the treatment includes XL765 (Exelixis) with erlotinib (NCT00777699), letrozole (NCT01082068), and temozolomide (NCT00704080) in non-small cell lung cancer, breast cancer, and gliomas, respectively.

Both BEZ235 and XL765 have shown good tolerability, with adverse effects including diarrhea, anorexia, and nausea [49]. Furthermore, the combined therapy using rapamycin and dual PI3K/mTOR kinase inhibitor (PI-103) has been shown to be efficacious against human ovarian cells *in vivo* [183].

Inhibitor	Trade name (company)	Drug target	Development stage	Tumor types	Reference
LY294002	-	Pan-PI3K inhibitor	Preclinical	-	[57,58]
Wortmannin	-	Pan-PI3K inhibitor	Preclinical	-	[57,59,60]
PX-866	(Oncothyreon)	Pan-PI3K inhibitor	Phase II	Solid cancers, prostate, colorectal, glioblastoma, SCCHN, non-small cell lung cancer	[62,64]
NVP-BKM120	Buparlisib (Novartis)	Pan-PI3K inhibitor	Phase III	Non-small cell lung cancer, prostate, breast, GBM, colon	[65,66]

Inhibitor	Trade name (company)	Drug target	Development stage	Tumor types	Reference
XL147	Pilaralisib (Sanofi-Exelixis)	Pan-PI3K inhibitor	Phase II	Solid cancers, breast, breast, endometrial, ovarian, non-small cell lung cancer, glioblastoma, lymphoma	[69,72]
GDC-0941	Pictilisib (Genentech-Roche)	Pan-PI3K inhibitor	Phase II	Solid cancers, breast, non-small cell lung cancer, glioblastoma, non-Hodgkin's lymphoma	[75,81]
GSK-2636771	(GlaxoSmithKline)	PI3K β inhibitor	Phase I	Solid cancers (PTEN deficient), prostate	[84]
NVP-BYL719	Alpelisib (Novartis)	PI3K α inhibitor	Phase II	Advanced solid tumors, SCCHN, breast, ovarian	[87,90]
GDC-0032	Taselisib (Genentech)	PI3K α inhibitor	Phase III	Solid cancers, breast, non-small cell lung cancer	[91,93]
CAL-101	Idelalisib (Gilead Sciences)	PI3K δ inhibitor	Phase III	Lymphomas, multiple myelomas, chronic lymphocytic leukemia, acute myeloid leukemia	[94,97,98]
KRX-0401	Perifosine (Pfizer)	AKT inhibitors	Phase II	Solid tumors, non-small cell lung cancer, colon, kidney, breast, gliomas, multiple myeloma, leukemia, lymphomas	[107,111,112,113]
GSK-690693	(GlaxoSmithKline)	ATP-competitive AKT inhibitor	Phase I	Hematologic malignancies	[114]
Rapamycin	Sirolimus (Wyeth)	Inhibits mTOR kinase by binding to FKBP12	Phase I	Glioblastoma, non-small cell lung cancer	[182]

Inhibitor	Trade name (company)	Drug target	Development stage	Tumor types	Reference
RAD001	Everolimus (Novartis)	Inhibits mTOR kinase by binding to FKBP12	Phase I/II/III (FDA has approved for RCC, 2009)	Metastatic renal cell carcinoma, breast cancer, melanoma, ovarian cancer, neuroendocrine tumors of the pancreatic origin (PNET), endometrial carcinoma	[56,133,134,135,136], NCT01939418, NCT01931163
CCI-779	Temsirolimus (Wyeth/Pfizer)	Inhibits mTOR kinase by binding to FKBP12	Phase I/II/III (FDA and European Medicine Agency have approved for RCC, 2007)	Non-small cell lung cancer; advanced solid tumors, metastatic renal cell carcinoma, hepatocellular carcinoma, cervical cancer, clear cell adenocarcinoma	[138,139,141]
MK-8669/AP23573	Ridaforolimus	Inhibits mTOR kinase by binding to FKBP12	Phase I/II/III	Sarcoma, bone, endometrial cancer	[144,145]
PP242		ATP competitive inhibitor of mTOR	Studies <i>in vitro</i> and <i>in vivo</i>	Colon cancer, acute myeloid leukemia	[156]
Torin2		ATP competitive inhibitor of mTOR	Studies <i>in vitro</i> and <i>in vivo</i>	Lung cancer	[159]
AZD8055		ATP competitive inhibitor of mTOR	Phase I	Advanced solid tumors, lymphoma	[183,184]
OSI-027		ATP competitive inhibitor of mTOR	Phase I	Advanced solid tumors, lymphoma	[185]
INK128		ATP competitive inhibitor of mTOR	Phase I	Glioblastoma, advanced solid tumors.	NCT02142803
GSK795		ATP competitive inhibitor of mTOR	Phase I	Advanced solid tumors	[134]
NVP-BEZ235	(Novartis)	Dual mTOR/PI3K	Phase I/II	Advanced solid tumors, breast cancer, prostate cancer	[94], NCT00620594

Inhibitor	Trade name (company)	Drug target	Development stage	Tumor types	Reference
BGT226	(Novartis)	Dual mTOR/PI3K	Phase I	Advanced solid tumors, breast cancer	[169], NCT00600275, NCT00742105
GDC-0980	(Genentech)	Dual mTOR/PI3K	Phase I/II	Non-Hodgkin lymphoma, endometriose	NCT00854126, NCT00854152, NCT01455493
PF-04691502	(Pfizer)	Dual mTOR/PI3K	Phase I	Advanced solid tumors	NCT00927823
GSK2126458	GlaxoSmithKline	Dual mTOR/PI3K	Phase I	Advanced solid tumors	NCT00972686, NCT01248858
XL765	(Exelixis)	Dual mTOR/PI3K	Phase I/II	Non-small cell lung cancer, breast cancer, gliomas	NCT00777699, NCT01082068, NCT00704080

Table 2. Overview of PI3K/AKT/mTOR pathway inhibitors.

9. Conclusions/future perspectives

Advances in molecular research have resulted in an improved understanding of cancer biology. There is strong preclinical rationale to support the continued development of PI3K/AKT/mTOR inhibitors, especially in some genetically defined cancer subtypes that may be the most sensitive to single-agent PI3K pathway inhibitors. These include cancers with PIK3CA activating mutations, mutations in PIK3R (p85 subunit), mutations or amplifications in one of the AKT isoforms or loss of PTEN. However, rational clinical trials design with a focus in identifying a patient population most likely to benefit from this strategy is imperative to the success of single-agent therapeutics.

The combination of PI3K/AKT/mTOR inhibitors with cytotoxic chemotherapy and other biological agents such as anti-HER2 compounds, EGFR inhibitors, and antiangiogenic agents may optimize the action of those agents in different pathways that control protein translation, cell growth, migration, metastasis, and angiogenesis. The successful development of the combinations will require determining the duration, doses, and schedules of targeted therapy and how to best incorporate it into standard treatment protocols. Several clinical trials are underway to prove the clinical use of the PI3K/AKT/mTOR inhibitors. The druggability of the components of the PI3K/AKT/mTOR signaling cascade, in addition to the enlightenment of the mutational landscape of human cancers, which points to the high frequency of genetic alterations and anomalous activation of the pathway, strongly suggests that targeting its elements might represent a useful treatment strategy in the fight against cancer.

Author details

Isabella S. Guimarães^{1,2}, Nayara G. Tessarollo^{1,2}, Paulo C.M. Lyra-Júnior^{1,2},
Diandra Z. dos Santos^{1,3}, Roger C. Zampier¹, Laura F.R.L. de Oliveira¹, Krislayne V. Siqueira¹,
Ian V. Silva^{2,4} and Leticia B.A. Rangel^{1,2,3*}

*Address all correspondence to: lbarangel@yahoo.com

1 Laboratory of Cellular and Molecular Biology of Human Cancer, Department of
Pharmaceutical Sciences, Federal University of Espirito Santo State, Brazil

2 Biotechnology Program, Federal University of Espirito Santo State, Brazil

3 Biochemistry and Pharmacology Program, Federal University of Espirito Santo State,
Brazil

4 Department of Morphology, Health Sciences Center, Federal University of Espirito Santo
State, Brazil

References

- [1] Gomez-Pinillos A., Ferrari A.C. mTOR signaling pathway and mTOR inhibitors in cancer therapy. *Hematology/Oncology Clinics of North America*. 2012;26(3):483-505. DOI: 10.1016/j.hoc.2012.02.014
- [2] Yuan T.L., Cantley L.C. PI3K pathway alterations in cancer: variations on a theme. *Oncogene*. 2008;27:5497–5510. DOI: 10.1038/onc.2008.245
- [3] Li T., Wang G. Computer-aided targeting of the PI3K/Akt/mTOR pathway: toxicity reduction and therapeutic opportunities. *International Journal of Molecular Sciences*. 2014;15(10):18856-91. DOI: 10.3390/ijms151018856
- [4] Bartholomeusz C., Gonzalez-Angulo, A.M. Targeting the PI3K signaling pathway in cancer therapy. *Expert Opinion on Therapeutic Targets*. 2012;16(1):121-30. DOI: 10.1517/14728222.2011.644788
- [5] Kandoth C., McLellan M.D., Vandin F., Ye K., Niu B., Lu C., et al. Mutational landscape and significance across 12 major cancer types. *Nature*. 2013;502(7471):333-339. DOI: 10.1038/nature12634
- [6] Vanhaesebroeck B., Guillermet-Guibert J., Graupera M., Bilanges B. The emerging mechanisms of isoform-specific PI3K signalling. *Nature Reviews Molecular Cell Biology*. 2010a;11 (5):329-341. DOI: 10.1038/nrm2882

- [7] Vadas O., Burke J. E., Zhang X., Berndt A., Williams R. L. Structural basis for activation and inhibition of class I phosphoinositide 3-kinases. *Science Signaling*. 2011;4 (195):re2. DOI: 10.1126/scisignal.2002165
- [8] Hawkins P.T., Anderson K.E, Davidson K., Stephens L.R. Signalling through class I PI3Ks in mammalian cells. *Biochemical Society Transactions*. 2006;34(5):647–662. DOI: 10.1042/BST0340647
- [9] Patrucco E., Notte A., Barberis L., Selvetella G., Maffei A., Brancaccio M., et al. PI3Kgamma modulates the cardiac response to chronic pressure overload by distinct kinase-dependent and -independent effects. *Cell*. 2004;118(3): 375-387. DOI: 10.1016/j.cell.2004.07.017
- [10] Hirsch E., Braccini L., Ciraolo E., Morello F., Perino A. Twice upon a time: PI3K's secret double life exposed. *Trends in Biochemical Sciences*. 2009;34(5): 244- 248. DOI: 10.1016/j.tibs.2009.02.003
- [11] Dou Z., Chattopadhyay M., Pan J.A., Guerriero J.L., Jiang, Y. P., Ballou L.M., et al. The class IA phosphatidylinositol 3-kinase p110-beta subunit is a positive regulator of autophagy. *Journal of Cell Biology*. 2010;191 (4): 827-843. DOI: 10.1083/jcb.201006056
- [12] Rauch J., Volinsky N., Romano D., Kolch W. The secret life of kinases: functions beyond catalysis. *Cell Communication and Signaling*. 2011;9(1):23. DOI: 10.1186/1478-811X-9-23
- [13] Dou Z., Pan J.A., Dbouk H.A., Ballou L.M., DeLeon J.L., Fan Y., et al. Class IA PI3K p110b subunit promotes autophagy through Rab5 small GTPase in response to growth factor limitation. *Molecular Cell*. 2013;50 (1): 29-42. DOI: 10.1016/j.molcel.2013.01.022
- [14] Vanhaesebroeck B., Vogt P.K., Rommel C. PI3K: from the bench to the clinic and back. *Current Topics in Microbiology and Immunology*. 2010b;347:1-19. DOI: 10.1007/82_2010_65
- [15] Gaullier J.M., Simonsen A., D'Arrigo A., Bremnes B., Stenmark H, Aasland R. FYVE fingers bind PtdIns (3)P. *Nature*. 1998;394(6692):432-433.DOI: 10.1038/28767
- [16] Mu F.T., Callaghan J.M., Steele-Mortimer O., Stenmark H., Parton R.G., Campbell P.L., et al. EEA1, an early endosome-associated protein. EEA1 is a conserved alpha-helical peripheral membrane protein flanked by cysteine "fingers" and contains a calmodulin-binding IQ motif. *Journal of Biological Chemistry*. 1995;270 (22):13503–13511.DOI: 10.1074/jbc.270.22.13503
- [17] Stenmark H., Aasland R., Toh B.H., D'Arrigo A. Endosomal localization of the auto-antigen EEA1 is mediated by a zinc-binding FYVE finger. *Journal of Biological Chemistry*. 1996;271 (39):24048–24054. DOI: 10.1074/jbc.271.39.24048

- [18] Harlan J.E., Hajduk P.J., Yoon H.S., Fesik S.W. Pleckstrin homology domains bind to phosphatidylinositol-4,5-bisphosphate. *Nature*. 1994;371(6493):168–170. DOI: 10.1038/371168a0
- [19] Cheever M.L., Sato T.K., de Beer T., Kutateladze T.G, Emr S.D., Overduin M. Phox domain interaction with PtdIns(3)P targets the Vam7 t-SNARE to vacuole membranes. *Nature Cell Biology*. 2001;3(7):613–8. DOI: 10.1038/35083000
- [20] Ellson C.D., Gobert-Gosse S., Anderson K.E., Davidson K, Erdjument-Bromage H., Tempst P., et al. PtdIns(3)P regulates the neutrophil oxidase complex by binding to the PX domain of p40(phox). *Nature Cell Biology*. 2001;3(7):679-682. DOI: 10.1038/35083076
- [21] Kanai F., Liu H., Field S.J., Akbary H., Matsuo T., Brown G.E., et al. The PX domains of p47phox and p40phox bind to lipid products of PI(3)K. *Nature Cell Biology*. 2001;3(7):675-678. DOI: 10.1038/35083070
- [22] Song X., Xu W., Zhang A., Huang G., Liang X., Virbasius J.V., et al. Phox homology domains specifically bind phosphatidylinositol phosphates. *Biochemistry*. 2001;40(30):8940-8944. DOI: 10.1021/bi0155100
- [23] Xu Y., Hortsman H., Seet L., Wong S.H., Hong W. SNX3 regulates endosomal function through its PXdomain-mediated interaction with PtdIns(3)P. *Nature Cell Biology*. 2001;3(7):658-666. DOI: 10.1038/35083051
- [24] Sarbassov D.D., Ali S.M., Sengupta S., Sheen J.H., Hsu P.P., Bagley A.F., et al. Prolonged rapamycin treatment inhibits mTORC2 assembly and Akt/PKB. *Molecular Cell*. 2006;22(2):159-168. DOI: <http://dx.doi.org/10.1016/j.molcel.2006.03.029>
- [25] Petrulea M.S., Plantinga T.S., Smit J.W., Georgescu C.E., Netea-Maier R.T. PI3K/Akt/mTOR: a promising therapeutic target for non-medullary thyroid carcinoma. *Cancer Treatment Reviews*. 2015;26. DOI: 10.1016/j.ctrv.2015.06.005
- [26] Mukohara T. PI3K mutations in breast cancer: prognostic and therapeutic implications. *Breast Cancer* (Dove Medical Press). 2015;7:111-23. DOI: 10.2147/BCTT.S60696
- [27] Hay N, Sonenberg N. Upstream and downstream of mTOR. *Genes & Development*. 2004;18(16):1926-45. DOI: 10.1101/gad.1212704
- [28] Larue L., Bellacosa A. Epithelial–mesenchymal transition in development and cancer: role of phosphatidylinositol 3' kinase/AKT pathways. *Oncogene*. 2005;24(50):7443-7454. DOI: 10.1038/sj.onc.1209091
- [29] Manning B.D., Cantley L.C. Rheb fills a GAP between TSC and TOR. *Trends of Biochemical Sciences*. 2003;28(11):573-6. DOI: <http://dx.doi.org/10.1016/j.tibs.2003.09.003>
- [30] Engelman J.A., Luo J., Cantley L.C. The evolution of phosphatidylinositol 3-kinases as regulators of growth and metabolism. *Nature Reviews Genetics*. 2006;7(8):606-19. DOI: 10.1038/nrg1879

- [31] Jacinto E., Loewith R., Schmidt A., Lin S., Ruegg M.A., Hall A., et al. Mammalian TOR complex 2 controls the actin cytoskeleton and is rapamycin insensitive. *Nature Cell Biology*. 2004;6:1122–1128. DOI: 10.1038/ncb1183
- [32] Vadlakonda L., Dash A., Pasupuleti M., Anil K. K., Reddanna P. The paradox of Akt-mTOR interactions. *Frontiers in Oncology*. 2013;3(165):1-9. DOI: 10.3389/fonc.2013.00165.
- [33] Yin Y., Shen W.H. PTEN: a new guardian of the genome. *Oncogene*. 2008;27 (41): 5443-5453. DOI: 10.1038/onc.2008.241
- [34] Chiang G.G., Abraham R.T. Targeting the mTOR signaling network in cancer. *Trends in Molecular Medicine*. 2007;13(10):433-42. DOI : <http://dx.doi.org/10.1016/j.molmed.2007.08.001>
- [35] Leary A., Auclin E., Pautier P., Lhommé C. The PI3K/Akt/mTOR pathway in ovarian cancer: biological rationale and therapeutic opportunities. *Ovarian Cancer—A Clinical and Translational Update*. 2013;13:275-302. DOI: 10.5772/54170
- [36] Stemke-Hale K., Gonzalez-Angulo A.M., Lluch A., Neve R.M., Kuo W.L., Davies M., et al. An integrative genomic and proteomic analysis of PIK3CA, PTEN, and AKT mutations in breast cancer. *Cancer Research*. 2008;68(15):6084-6091. DOI: 10.1158/0008-5472.CAN-07-6854
- [37] Fresno V.J.A, Casado E., de Castro J., Cejas P., Belda-Iniesta C., González-Barón M. PI3K/Akt signalling pathway and cancer. *Cancer Treatment Reviews*. 2004;30(2): 193-204. DOI: 10.1016/j.ctrv.2003.07.007
- [38] Tang J.M., He Q.Y., Guo R.X., Chang X.J. Phosphorylated Akt overexpression and loss of PTEN expression in non-small cell lung cancer confers poor prognosis. *Lung Cancer*. 2006;51(2):181-191. DOI: 10.1016/j.lungcan.2005.10.003
- [39] Cheng H., Shcherba M., Pendurti G., Liang Y., Piperdi B., Perez-Soler R. Targeting the PI3K/AKT/mTOR pathway: potential for lung cancer treatment. *Lung Cancer Management*. 2014;3(1):67-75. DOI: 10.2217/lmt.13.72
- [40] Bellacosa A., de Feo D., Godwin A.K., Bell D.W., Cheng J.Q., Altomare D.A., et al. Molecular alterations of the AKT2 oncogene in ovarian and breast carcinomas. *International Journal of Cancer*. 1995;64(4):280-5. DOI: 10.1002/ijc.2910640412
- [41] Shayesteh L., Lu Y., Kuo W.L., Baldocchi R., Godfrey T., Collins C., et al. PIK3CA is implicated as an oncogene in ovarian cancer. *Nature Genetics*. 1999;21:99-102. DOI: 10.1038/5042
- [42] Campbell I.G., Russell S.E., Choong D.Y., Montgomery K.G., Ciavarella M.L., Hooi C.S. et al. Mutation of the PIK3CA gene in ovarian and breast cancer. *Cancer Research*. 2004;64:7678-81. DOI: 10.1158/0008-5472.CAN-04-2933

- [43] Kuo K.T, Mao T. L., Jones S., Veras E., Ayhan A., Wang T.L., et al. Frequent activating mutations of PIK3CA in ovarian clear cell carcinoma. *American Journal of Pathology*. 2009;174(5): 1597–1601. DOI: 10.2353/ajpath.2009.081000
- [44] Kinross K.M., Montgomery K.G., Kleinschmidt M., Waring P., Ivetac I., Tikoo A. et al. An activating Pik3ca mutation coupled with Pten loss is sufficient to initiate ovarian tumorigenesis in mice. *Journal of Clinical Investigation*. 2012;122(2):553-557. DOI: 10.1172/JCI59309
- [45] Tanwar P.S., Zhang L., Kaneko-Tarui T., Curley M.D., Taketo M.M., Rani P. Mammalian target of rapamycin is a therapeutic target for murine ovarian endometrioid adenocarcinomas with dysregulated Wnt/ β -catenin and PTEN. *PLoS One*. 2011;6(6):e20715. DOI: 10.1371/journal.pone.0020715
- [46] Steuer-Vogt M.K., Bonkowsky V., Ambrosch P., Scholz M., Neiss A., Strutz J., et al. The effect of an adjuvant mistletoe treatment programme in resected head and neck cancer patients: a randomised controlled clinical trial. *European Journal of Cancer*. 2001;37(1):23-31. DOI: 10.1016/S0959-8049(00)00360-9
- [47] Dancey J.E. Clinical development of mammalian target of rapamycin inhibitors. *Hematology Oncology Clinics North of American*. 2002;16(5):1101-14. PII: S0889-8588(02)00051-5
- [48] Huang S., Houghton P.J. Inhibitors of mammalian target of rapamycin as novel anti-tumor agents: from bench to clinic. *Current Opinion in Investigational Drugs*. 2002;3(2):295-304. PMID: 12020063
- [49] Wander S.A, Hennessy B.T, Slingerland J.M. Next-generation mTOR inhibitors in clinical oncology: how pathway complexity informs therapeutic strategy. *Journal of Clinical Investigation*. 2011;121(4):1231-1241. DOI: 10.1172/JCI44145
- [50] Mabuchi S., Kawase C., Altomare D.A., Morishige K., Sawada K., Hayashi M., et al. mTOR is a promising therapeutic target both in cisplatin-sensitive and cisplatin-resistant clear cell carcinoma of the ovary. *Clinical Cancer Research*. 2009;15 (17): 5404-5413. DOI: 10.1158/1078-0432.CCR-09-0365
- [51] Peng C.L., Lai P.S., Lin F.H., Yueh-Hsiu W.S., Shieh M.J. Dual chemotherapy and photodynamic therapy in an HT-29 human colon cancer xenograft model using SN-38-loaded chlorin-core star block copolymer micelles. *Biomaterial*. 2009;30:3614–3625. DOI: 10.1016/j.biomaterials.2009.03.048
- [52] Zhang, P.; Hu, L.; Yin, Q.; Zhang, Z.; Feng, L.; Li, Y. Transferrin-conjugated polyphosphoester hybrid micelle loading paclitaxel for brain-targeting delivery: synthesis, preparation and in vivo evaluation. *Journal of Controlled Release* 2012;159:429–434. DOI: 10.1016/j.jconrel.2012.01.031

- [53] Rodon J., Dienstmann R., Serra V., Tabernero J. Development of PI3K inhibitors: lessons learned from early clinical trials. *Nature Reviews. Clinical Oncology*. 2013;10(3): 143–153. DOI: 10.1038/nrclinonc.2013.10
- [54] Yao J.C, Shah M.H., Ito T., Bohas C.L., Wolin E.M., Van Cutsem E., et al. RAD001 in Advanced Neuroendocrine Tumors, Third Trial (RADIANT-3) Study Group. Everolimus for advanced pancreatic neuroendocrine tumors. *New England Journal of Medicine*. 2013;364(6):514–523. DOI: 10.1056/NEJMoa1009290.
- [55] Hudes G., Carducci M., Tomczak P., Dutcher J., Figlin R., Kapoor A., et al. Temsirolimus, interferon alfa, or both for advanced renal-cell carcinoma. *New England Journal of Medicine*. 2007;356(22):2271–2281. DOI: 10.1056/NEJMoa066838
- [56] Baselga J., Campone M., Piccart M., Burris H.A., Rugo H.S., Sahmoud T., et al. Everolimus in postmenopausal hormone-receptor-positive advanced breast cancer. *New England Journal of Medicine*. 2012;366(6):520–529. DOI: 10.1056/NEJMoa1109653
- [57] Davies S.P., Reddy H., Caivano M., Cohen P. Specificity and mechanism of action of some commonly used protein kinase inhibitors. *Biochemical Journal*. 2000;351(1): 95–105.
- [58] Vlahos C.J., Matter W.F., Hui K.Y., Brown R.F. A specific inhibitor of phosphatidylinositol 3-kinase, 2-(4-morpholinyl)-8-phenyl-4H-1-benzopyran-4-one (LY294002). *Journal of Biological Chemistry*. 1994;269(7): 5241–5248.
- [59] Knight Z.A., Shokat K.M. Chemically targeting the PI3K family. *Biochemical Society Transactions*. 2007;35(2):245–249. DOI: 10.1042/BST0350245
- [60] Marone R., Cmiljanovic V., Giese B., Wymann M.P. Targeting phosphoinositide 3-kinase-moving towards therapy. *Biochimica et Biophysica Acta*. 2008;1784(1):159–185. DOI: 10.1016/j.bbapap.2007.10.003
- [61] Garcia-Echeverria C., Sellers W.R. Drug discovery approaches targeting the PI3K/Akt pathway in cancer. *Oncogene*. 2008;27(41):5511–5526. DOI: 10.1038/onc.2008.246
- [62] Ihle N.T, Williams R., Chow S., Chew W., Berggren M.I., Paine-Murrieta G., et al. Molecular pharmacology and antitumor activity of PX-866, a novel inhibitor of phosphoinositide-3-kinase signaling. *Molecular Cancer Therapeutics*. 2004;3(7):763–72.
- [63] Ihle N.T., Paine-Murrieta G., Berggren M.I, Baker A., Tate W.R, Wipf P., et al. The phosphatidylinositol-3-kinase inhibitor PX-866 overcomes resistance to the epidermal growth factor receptor inhibitor gefitinib in A-549 human non-small cell lung cancer xenografts. *Molecular Cancer Therapeutics*. 2005;4(9):1349–1357. DOI: 10.1158/1535-7163.MCT-05-0149
- [64] Hong D.S., Bowles D.W., Falchook G.S., Messersmith W.A., George G.C., O'Bryant C.L., et al. A multicenter phase I trial of PX-866, an oral irreversible phosphatidylinositol 3-kinase inhibitor, in patients with advanced solid tumors. *Clinical Cancer Research*. 2012;18(15):4173–82. DOI: 10.1158/1078-0432.CCR-12-0714

- [65] Burger M., Pecchi S., Wagman A., Ni Z.J., Knapp M., Hendrickson T., et al. Identification of NVP-BKM120 as a potent, selective, orally bioavailable class I PI3 kinase inhibitor for treating cancer. *ACS Medicinal Chemistry Letters*. 2011;2(10):774-779. DOI: 10.1021/ml200156t
- [66] Maira S.M., Pecchi S., Huang A., Burger M., Knapp M., Sterker D., et al. Identification and characterization of NVP-BKM120, an orally available pan-class I PI3-kinase inhibitor. *Molecular Cancer Therapeutics*. 2012;11(2):317-328. DOI: 10.1158/1535-7163.MCT-11-0474.
- [67] Bendell J.C., Rodon J., Burris H.A., de Jonge M., Verweij J., Birle D., et al. Phase I, dose-escalation study of BKM120, an oral pan-Class I PI3K inhibitor, in patients with advanced solid tumors. *Journal of Clinical Oncology*. 2012;30(3):282-290. DOI: 10.1200/JCO.2011.36.1360
- [68] Rodon J., Braña I., Siu L.L., De Jonge M.J., Homji N., Mills D., et al. Phase I dose-escalation and -expansion study of buparlisib (BKM120), an oral pan-Class I PI3K inhibitor, in patients with advanced solid tumors. *Investigational New Drugs*. 2014;32(4):670-81. DOI: 10.1007/s10637-014-0082-9
- [69] Edelman G., Bedell C., Shapiro G., Pandya S.S., Kwak E.L., Scheffold C., et al. A phase I dose-escalation study of XL147 (SAR245408), a PI3K inhibitor administered orally to patients (pts) with advanced malignancies. *Journal of Clinical Oncology*. 2010;28(15s):3004.
- [70] Markman B., Dienstmann R., Tabernero J. Targeting the PI3K/Akt/mTOR pathway—beyond rapalogs. *Oncotarget*. 2010;1(7):530-543. PMC3248125
- [71] Foster P., Yamaguchi K., Hsu P.P., Qian F., Du X., Wu J., et al. The selective PI3K inhibitor XL147 (SAR245408) inhibits tumor growth and survival and potentiates the activity of chemotherapeutic agents in preclinical tumor models. *Molecular Cancer Therapeutics*. 2015;14(4):931-940. DOI: 10.1158/1535-7163.MCT-14-0833
- [72] Shapiro G.I., Rodon J., Bedell C., Kwak E.L., Baselga J., Braña I., et al. Phase I safety, pharmacokinetic, and pharmacodynamic study of SAR245408 (XL147), an oral pan-class I PI3K inhibitor, in patients with advanced solid tumors. *Clinical Cancer Research*. 2014;20(1):233-45. DOI: 10.1158/1078-0432.CCR-13-1777
- [73] Soria J.C., LoRusso P., Bahleda R., Lager J., Liu L., Jiang J., et al. Phase I dose-escalation study of pilaralisib (SAR245408, XL147), a pan-Class I PI3K inhibitor, in combination with erlotinib in patients with solid tumors. *Oncologist*. 2015;20(3):245-6. DOI: 10.1634/theoncologist.2014-0449
- [74] Tolaney S., Burris H., Gartner E., Mayer I.A., Saura C., Maurer M., et al. Phase I/II study of pilaralisib (SAR245408) in combination with trastuzumab or trastuzumab plus paclitaxel in trastuzumab-refractory HER2-positive metastatic breast cancer. *Breast Cancer Research Treatment*. 2015;149(1):151-161. DOI: 10.1007/s10549-014-3248-4

- [75] Folkes A., Ahmadi K., Alderton W.K., Alix S., Baker S.J., Box G. et al. The identification of 2-(1H-indazol-4-yl)-6-(4-methanesulfonyl-piperazin-1-ylmethyl)-4-morpholin-4-yl-1H-hieno[3,2-d]pyrimidine (GDC-0941) as a potent, selective, orally bioavailable inhibitor of class I PI3 kinase for the treatment of cancer. *Journal Medicinal Chemistry*. 2008;51(18):5522-5532. DOI: 10.1021/jm800295d
- [76] Sampath D., Belvin M., Guan J., Edgar K., Wallin J., Prior W.W., et al. Combination of class I PI3K inhibitor, GDC-0941, with standard of care therapeutics results in enhanced anti-tumor responses in human cancer models in vitro and in vivo. *European Journal of Cancer Supplements*. 2008;6(12):69-70. DOI: 10.1016/S1359-6349(08)72152-9
- [77] Zou Z.Q., Zhang L.N., Wang F., Bellenger J, Shen Y.Z., Zhang X.H. The novel dual PI3K/mTOR inhibitor GDC-0941 synergizes with the MEK inhibitor U0126 in non-small cell lung cancer cells. *Molecular Medicine Reports*. 2012;5(2):503-8. DOI: 10.3892/mmr.2011.682
- [78] Wallin J.J., Guan J., Prior W.W, Lee L.B, Berry L., Belmont L.D., Koeppen H., Belvin M., Friedman L.S., Sampath D. GDC-0941, a novel class I selective PI3K inhibitor, enhances the efficacy of docetaxel in human breast cancer models by increasing cell death in vitro and in vivo. *Clinical Cancer Research*. 2012;18(14):3901-11. DOI: 10.1158/1078-0432.CCR-11-2088
- [79] Burrows N., Babur M., Resch J., Ridsdale S., Mejin M., Rowling E.J., et al. GDC-0941 inhibits metastatic characteristics of thyroid carcinomas by targeting both the phosphoinositide-3 kinase (PI3K) and hypoxia-inducible factor-1 α (HIF-1 α) pathways. *Journal of Clinical Endocrinology and Metabolism*. 2011;96(12):e1934-1943. DOI: 10.1210/jc.2011-1426
- [80] Kwei K.A., Baker J.B., Pelham R.J. Modulators of sensitivity and resistance to inhibition of PI3K identified in a pharmacogenomic screen of the NCI-60 human tumor cell line collection. *PLoS One*. 2012;7(9):e46518. DOI: 10.1371/journal.pone.0046518
- [81] Sarker D., Ang J.E., Baird R., Kristeleit R., Shah K., Moreno V., et al. First-in-human phase I study of pictilisib (GDC-0941), a potent pan-Class I phosphatidylinositol-3-kinase (PI3K) inhibitor, in patients with advanced solid tumors. *Clinical Cancer Research*. 2015;21(1):77-86. DOI: 10.1158/1078-0432.CCR-14-0947
- [82] Jia S., Liu Z., Zhang S., Liu P., Zhang L., Lee S.H., et al. Essential roles of PI(3)K-p110[bgr] in cell growth, metabolism and tumorigenesis. *Nature*. 2008;454:776-779. DOI: 10.1038/nature07091
- [83] Wee S., Wiederschain D., Maira S.M., Loo A., Miller C., deBeaumont R., et al. PTEN-deficient cancers depend on PIK3CB. *Proceedings of the National Academy of Sciences*. 2008;105:13057-13062. DOI: 10.1073/pnas.0802655105
- [84] Ni J., Liu Q., Xie S., Carlson C., Von T., Vogel K., et al. Functional characterization of an isoform-selective inhibitor of PI3K-p110 β as a potential anticancer agent. *Cancer Discovery*. 2012;2(5):425-433. DOI: 10.1158/2159-8290.CD-12-0003

- [85] Rommel C., Camps M., Ji H. PI3K delta and PI3K gamma: partners in crime in inflammation in rheumatoid arthritis and beyond? *Nature Reviews Immunology*. 2007;7(3):191-201. DOI: 10.1038/nri2036
- [86] U.S. Food and Drug Administration. U.S. Department of Health and Human Services. [Internet]. 2015. Available from: <http://www.fda.gov/ICECI/Inspections/ucm250729.htm> [Accessed: 2015-03-11]
- [87] Fritsch C., Huang A., Chatenay-Rivauday C., Schnell C., Reddy A., Liu M., et al. Characterization of the novel and specific PI3K α inhibitor NVP-BYL719 and development of the patient stratification strategy for clinical trials. *Molecular Cancer Therapeutics*. 2014;13(5):1117-29. DOI: 10.1158/1535-7163.MCT-13-0865
- [88] Fritsch C.M., Schnell C., Chatenay-Rivauday C., Guthy D.A., de Pover A., Wartmann M., et al. NVP-BYL719, a novel PI3K α selective inhibitor with all the characteristics required for clinical development as an anti-cancer agent. *Cancer Research*. 2012;3748. DOI: 10.1158/1538-7445.AM2012-3748
- [89] Huang A., Fritsch C., Wilson C., et al. Single agent activity of PIK3CA inhibitor BYL719 in a broad cancer cell line panel. *Cancer Research*. 2012;3749. DOI: 10.1158/1538-7445.AM2012-3749
- [90] Gonzalez-Angulo A.M., Juric D., Argiles G., Schellens J.H., Burris H.A, Berlin J., et al. Safety, pharmacokinetics, and preliminary activity of the α -specific PI3K inhibitor BYL719: results from the first-in-human study. *Journal of Clinical Oncology*. 2013;31:Abstract: 2531
- [91] Ndubaku C.O., Heffron T.P., Staben S.T., Baumgardner M., Blaquiere N., Bradley E., et al. Discovery of 2-{3-[2-(1-isopropyl-3-methyl-1H-1,2,4-triazol-5-yl)-5,6 dihydrobenzo[f]imidazo[1,2-d][1,4]oxazepin-9-yl]-1H-pyrazol-1-yl}-2 methylpropanamide (GDC-0032): a β -sparing phosphoinositide 3-kinase inhibitor with high unbound exposure and robust in vivo antitumor activity. *Journal of Medicinal Chemistry*. 2013;56(11):4597-4610. DOI: 10.1021/jm4003632
- [92] Wallin J.J., Edgar K.A., Guan J., Sampath D., Nannini M., Belvin M., et al. Abstract P2-17-01: the PI3K inhibitor GDC-0032 is selectively potent against PIK3CA mutant breast cancer cell lines and tumors. *Cancer Research*. 2013;73:P2-17-01. DOI: 10.1158/0008-5472.SABCS13-P2-17-01
- [93] Juric D., Krop I., Ramanathan R.K., Xiao J., Sanabria S., Wilson T.R., et al. GDC-0032, a beta isoform-sparing PI3K inhibitor: results of a first-in-human phase Ia dose escalation study. In: AACR 104th Annual Meeting 2013; April 2013; Washington, DC; 73:LB-64.
- [94] Yang Q., Modi P., Newcomb T., Qu C., Gandhi V. Idelalisib: first-in-class PI3K delta inhibitor for the treatment of chronic lymphocytic leukemia, small lymphocytic leukemia, and follicular lymphoma. *Clinical Cancer Research*. 2015;21(7):1537-1542. DOI: 10.1158/1078-0432.CCR-14-2034

- [95] Herman S.E., Gordon A.L., Wagner A.J., Heerema N.A., Zhao W., Flynn J.M., et al. Phosphatidylinositol 3-kinase- δ inhibitor CAL-101 shows promising preclinical activity in chronic lymphocytic leukemia by antagonizing intrinsic and extrinsic cellular survival signals. *Blood*. 2010;116(12):2078-2088.
- [96] Lannutti B.J., Meadows S.A., Herman S.E., Kashishian A., Steiner B., Johnson A.J., et al. CAL-101, a p110 δ selective phosphatidylinositol-3-kinase inhibitor for the treatment of B-cell malignancies, inhibits PI3K signaling and cellular viability. *Blood*. 2011;117(2):591-594. DOI: 10.1182/blood-2010-03-275305
- [97] Webb H.K., Chen H., Yu A.S., et al. Clinical pharmacokinetics of CAL-101, a p110 δ isoform-selective PI3K inhibitor, following single- and multiple-dose administration in healthy volunteers and patients with hematological malignancies. *ISRN Oncology*. 2010;116(21):1774. DOI: 10.1155/2014/931858
- [98] Kahl B.S., Spurgeon S.E., Furman R.R., Flinn I.W., Coutre S.E., Brown J.R., et al. A phase 1 study of the PI3K δ inhibitor idelalisib in patients with relapsed/refractory mantle cell lymphoma (MCL). *Blood*. 2014;123(22):3398-3405. DOI: 10.1182/blood-2013-11-537555.
- [99] Kondapaka S.B., Singh S.S., Dasmahapatra G.P., Sausville E.A., Roy K.K. Perifosine, a novel alkylphospholipid, inhibits protein kinase B activation. *Molecular Cancer Therapeutics*. 2003;2(11):1093-1103.
- [100] Hilgard P., Klenner T., Stekar J., Nössner G., Kutscher B., Engel J. D-21266, a new heterocyclic alkylphospholipid with antitumour activity. *European Journal of Cancer*. 1997;33(3):442-446. DOI: 10.1016/S0959-8049(97)89020-X
- [101] Yao C., Wei J.J., Wang Z.Y., Ding H.M., Li D., Yan S.C., et al. Perifosine induces cell apoptosis in human osteosarcoma cells: new implication for osteosarcoma therapy? *Cell Biochemistry and Biophysics*. 2013;65(2):217-227. DOI: 10.1007/s12013-012-9423-5
- [102] Sun H., T. Yu, J. Li. Co-administration of perifosine with paclitaxel synergistically induces apoptosis in ovarian cancer cells: more than just AKT inhibition. *Cancer Letters*. 2011;310(1):118–128. DOI: 10.1016/j.canlet.2011.06.010
- [103] Engel J.B., Schönhals T., Häusler S., Krockenberger M., Schmidt M., Horn E., et al. Induction of programmed cell death by inhibition of AKT with the alkylphosphocholine perifosine in in vitro models of platinum sensitive and resistant ovarian cancers. *Archives of Gynecology and Obstetrics*. 2011;283(3):603-10. DOI: 10.1007/s00404-010-1457-6
- [104] Nyakern M., Cappellini A., Mantovani I, Martelli A.M. Synergistic induction of apoptosis in human leukemia T cells by the Akt inhibitor perifosine and etoposide through activation of intrinsic and Fas-mediated extrinsic cell death pathways. *Molecular Cancer Therapeutics*. 2006;5:1559-1570. DOI: 10.1158/1535-7163.MCT-06-0076
- [105] Hideshima T., Catley L., Yasui H., Ishitsuka K., Raje N., Mitsiades C., et al. Perifosine, an oral bioactive novel alkylphospholipid, inhibits Akt and induces in vitro and

in vivo cytotoxicity in human multiple myeloma cells. *Blood*. 2006;107(10):4053-4062. DOI: <http://dx.doi.org/10.1182/blood-2005-08-3434>

- [106] Xin Y., Shen X.D., Cheng L., Hong D.F., Chen B. Perifosine inhibits S6K1-Gli1 signaling and enhances gemcitabine-induced anti-pancreatic cancer efficiency. *Cancer Chemotherapy and Pharmacology*. 2014;73(4):711-719. DOI: 10.1007/s00280-014-2397-9
- [107] Posadas E.M, Gulley J., Arlen P.M., Trout A., Parnes H.L., Wright J., et al. A phase II study of perifosine in androgen independent prostate cancer. *Cancer Biology Therapy*. 2005;4(10):1133-1137. DOI: 10.4161/cbt.4.10.2064
- [108] Argiris A., Cohen E., Karrison T., Esparaz B., Mauer A., Ansari R., et al. Phase II trial of perifosine, an oral alkylphospholipid, in recurrent or metastatic head and neck cancer. *Cancer Biology and Therapy*. 2006;5(7):766-770. DOI: 10.4161/cbt.5.7.2874
- [109] Knowling M., Blackstein M., Tozer R., Bramwell V., Dancey J., Dore N., et al. A phase II study of perifosine (D-21226) in patients with previously untreated metastatic or locally advanced soft tissue sarcoma: a National Cancer Institute of Canada Clinical Trials Group trial. *Investigational New Drugs*. 2006;24(5):435-439. DOI: 10.1007/s10637-006-6406-7
- [110] Marsh Rde W., Rocha Lima C.M., Levy D.E., Mitchell E.P., Rowland Jr K.M., Benson A.B. A phase II trial of perifosine in locally advanced, unresectable, or metastatic pancreatic adenocarcinoma. *American Journal of Clinical Oncology*. 2007;30(1):26–31. DOI: 10.1097/01.coc.0000251235.46149.43
- [111] Leighl N.B., Dent S., Clemons M., Vandenberg T.A., Tozer R., Warr D.G., et al. A phase 2 study of perifosine in advanced or metastatic breast cancer. *Breast Cancer Research Treatment*. 2008;108(1):87-92. DOI: 10.1007/s10549-007-9584-x
- [112] Fu S., Hennessey B.T., Ng C.S., Ju Z., Coombes K.R., Wolf J.K., et al. Perifosine plus docetaxel in patients with platinum and taxane resistant or refractory high-grade epithelial ovarian cancer. *Gynecologic Oncology*. 2012;126(1):47-53. DOI: 10.1016/j.ygyno.2012.04.006
- [113] Guidetti A., Carlo-Stella C., Locatelli SL., Malorni W., Mortarini R., Viviani S., et al. Phase II study of perifosine and sorafenib dual-targeted therapy in patients with relapsed or refractory lymphoproliferative diseases. *Clinical Cancer Research*. 2014;20(22):5641-5651. DOI: 10.1158/1078-0432.CCR-14-0770
- [114] Rhodes N., Heerding D.A., Duckett D.R., Eberwein D.J., Knick V.B., Lansing T.J., et al. Characterization of an Akt kinase inhibitor with potent pharmacodynamic and antitumor activity. *Cancer Research*. 2008;68(7):2366-74. DOI: 10.1158/0008-5472.CAN-07-5783
- [115] Feng Z., Zhang H., Levine A. J., Jin S. The coordinate regulation of the p53 and mTOR pathways in cells. *PNAS, Proceedings of the National Academy of Sciences*. 2005;102(23): 8204–8209. DOI: 10.1073/pnas.0502857102

- [116] Guertin D.A., Sabatini D.M. The pharmacology of mTOR inhibition. *Science Signaling*. 2009;2(67):pe24. DOI: 10.1126/scisignal.267pe24
- [117] Vezina C., Kudelski A., Sehgal S.N. Rapamycin (AY-22,989), a new antifungal antibiotic. I. Taxonomy of the producing streptomycete and isolation of the active principle. *Journal of Antibiotics*. 1975;28(10):721-726. DOI: <http://doi.org/10.7164/antibiotics.28.721>
- [118] Singh K., Sun S., Vezina C. Rapamycin (AY-22,989), a new antifungal antibiotic. IV. Mechanism of action. *Journal of Antibiotics*. 1979;32(6):630-645. DOI: 10.7164/antibiotics.28.721
- [119] Tsang C. K., Qi H., Liu L. F., Zheng, X.F.S. Targeting mammalian target of rapamycin (mTOR) for health and diseases. *Drug Discovery Today*. 2007;12(3-4): 112-24. DOI: 10.1016/j.drudis.2006.12.008
- [120] Shafer A., Zhou C., Gehrig P.A., Boggess J.F., Bae-Jump VL. Rapamycin potentiates the effects of paclitaxel in endometrial cancer cells through inhibition of cell proliferation and induction of apoptosis. *International Journal of Cancer*. 2010;126(5): 1144-54. DOI: 10.1002/ijc.24837
- [121] Oldham S., Hafen E. Insulin/IGF and target of rapamycin signaling: a TOR de force in growth control. *Trends in Cell Biology*. 2003;13(2):79-85. DOI: [http://dx.doi.org/10.1016/S0962-8924\(02\)00042-9](http://dx.doi.org/10.1016/S0962-8924(02)00042-9)
- [122] Fang Y., Vilella-Bach M., Bachmann R., Flanigan A., Chen J. Phosphatidic acid-mediated mitogenic activation of mTOR signaling. *Science* 2001;294(5548):1942-1945. DOI: 10.1126/science.1066015
- [123] Hara K., Maruki Y., Long X., Yoshino K., Oshiro N., Hidayat S., et al. Raptor, a binding partner of target of rapamycin (TOR), mediates TOR action. *Cancer Cell*. 2002;110:177-189. DOI: [http://dx.doi.org/10.1016/S0092-8674\(02\)00833-4](http://dx.doi.org/10.1016/S0092-8674(02)00833-4)
- [124] Kim D.H., Sarbassov D.D., Ali S.M., King J.E., Latek R.R., Erdjument-Bromage H., et al. mTOR interacts with raptor to form a nutrient-sensitive complex that signals to the cell growth machinery. *Cancer Cell*. 2002;110:163-175. DOI: [http://dx.doi.org/10.1016/S0092-8674\(02\)00808-5](http://dx.doi.org/10.1016/S0092-8674(02)00808-5)
- [125] Liu Q., Chang J.W., Wang J., Kang S.A., Thoreen C.C., Markhard A., et al. Discovery of 1-(4-(4-propionylpiperazin-1-yl)-3-(trifluoromethyl)phenyl)-9-(quinolin-3-yl)benzo[h][1,6]naphthyridin-2(1H)-one as a highly potent, selective mammalian target of rapamycin (mTOR) inhibitor for the treatment of cancer. *Journal of Medicinal Chemistry*. 2010;53(19):7146-7155. DOI: 10.1021/jm101144f
- [126] Alvarado Y., Mita M.M., Vemulapalli S., Mahalingam D., Mita A.C. Clinical activity of mammalian target of rapamycin inhibitors in solid tumors. *Targeted Oncology*. 2011;6(2):69-94. DOI: 10.1007/s11523-011-0178-5

- [127] Zaytseva Y.Y., Valentino J.D., Gulhati P., Evers B.M. mTOR inhibitors in cancer therapy. *Cancer Letters*. 2012;319(1):1-7. DOI: 10.1016/j.canlet.2012.01.005
- [128] Brown E.J., Albers M.W., Shin T.B., Ichikawa K., Keith C.T., Lane W.S., et al. A mammalian protein targeted by G1-arresting rapamycin-receptor complex. *Nature*. 1994;369(6483):756-758. DOI: 10.1038/369756a0
- [129] Sabatini D.M., Erdjument-Bromage H., Lui M., Tempst P., Snyder S.H. RAFT1: a mammalian protein that binds to FKBP12 in a rapamycin-dependent fashion and is homologous to yeast TORs. *Cell*. 1994;78(1):35-43. DOI: [http://dx.doi.org/10.1016/0092-8674\(94\)90570-3](http://dx.doi.org/10.1016/0092-8674(94)90570-3)
- [130] Vignot S., Faivre S., Aguirre D., Raymond E. mTOR-targeted therapy of cancer with rapamycin derivatives. *Annals of Oncology*. 2005;16(4):524-37. DOI: 10.1093/annonc/mdi113
- [131] Duran I., Siu L.L., Oza A.M., Chung T.B., Sturgeon J., Townsley C.A., et al. Characterisation of the lung toxicity of the cell cycle inhibitor temsirolimus. *European Journal of Cancer*. 2006;42(12):1875-1880. DOI: <http://dx.doi.org/10.1016/j.ejca.2006.03.015>
- [132] National Cancer Institute [Internet]. 2013. Available from: <http://www.cancer.gov/about-cancer/treatment/drugs/fda-everolimus>. [Accessed: 2015-05-20].
- [133] Pavel M.E., Hainsworth J.D., Baudin E., Peeters M., Horsch D., Winkler R.E., et al. Everolimus plus octreotide long-acting repeatable for the treatment of advanced neuroendocrine tumours associated with carcinoid syndrome (RADIANT-2): a randomised, placebo-controlled, phase 3 study. *Lancet*. 2011;378(9808):2005-2012. DOI: 10.1016/S0140-6736(11)61742-X
- [134] Burris H.A., Lebrun F., Rugo H.S., Beck J.T, Piccart M., Neven P., et al. Health-related quality of life of patients with advanced breast cancer treated with everolimus plus exemestane versus placebo plus exemestane in the phase 3, randomized, controlled, BOLERO-2 trial. *Cancer*. 2013;119(10):1908-1915. DOI: 10.1002/cncr.28010
- [135] André F., O'Regan R., Ozguroglu M., Toi M., Xu B., Jerusalem G., et al. Everolimus for women with trastuzumab-resistant, HER2-positive, advanced breast cancer (BOLERO-3): a randomised, double-blind, placebo-controlled phase 3 trial. *Lancet Oncology*. 2014;15(6):580-91. DOI: 10.1016/S1470-2045(14)70138-X
- [136] Hurvitz S.A., Dalenc F., Campone M., O'Regan R.M., Tjan-Heijnen V.C., Gligorov J., et al. A phase 2 study of everolimus combined with trastuzumab and paclitaxel in patients with HER2-overexpressing advanced breast cancer that progressed during prior trastuzumab and taxane therapy. *Breast Cancer Research and Treatment*. 2013;141(3): 437-446. DOI: 10.1007/s10549-013-2689-5
- [137] Huang S., Houghton P.J. Targeting mTOR signaling for cancer therapy. *Current Opinion of Pharmacology* 2003;3:371-7. DOI: 10.1016/S1471-4892(03)00071-7

- [138] Bae-Jump V.L., Zhou C., Boggess J.F., Gehring P.A. Synergistic effect of rapamycin and cisplatin in endometrial cancer cells. *Cancer*. 2009;115:3887-96. DOI: 10.1002/cncr.24431
- [139] Temkin S.M., Fleming G. Current treatment of metastatic endometrial cancer. *Cancer Control*. 2009;16(1):38-45. PMID: 19078928
- [140] Tinker A.V., Ellard S., Welch S., Moens F., Allo G., Tsao M.S., et al. Phase II study of temsirolimus (CCI-779) in women with recurrent, unresectable, locally advanced or metastatic carcinoma of the cervix. A trial of the NCIC Clinical Trials Group (NCIC CTG IND 199). *Gynecologic Oncology* 2013;130:269–74. DOI: 10.1016/j.ygyno.2013.05.008
- [141] Takatori E., Shoji T., Miura Y., Takada A., Takeuchi S., Sugiyama T. Effective use of everolimus as salvage chemotherapy for ovarian clear cell carcinoma: a case report. *Onco Targets and Therapy*. 2014;7:165-169. DOI: 10.2147/OTT.S54745
- [142] Behbakht K., Sill M.W., Darcy K.M., Rubin S.C., Mannel R.S., Waggoner S., et al. Phase II trial of the mTOR inhibitor, temsirolimus and evaluation of circulating tumor cells and tumor biomarkers in persistent and recurrent epithelial ovarian and primary peritoneal malignancies: a Gynecologic Oncology Group study. *Gynecologic Oncology*. 2011;123(1):19-26. DOI: 10.1016/j.ygyno.2011.06.022
- [143] Zhang Y.J., Duan Y., Zheng X.F.S. Targeting the mTOR kinase domain: the second generation of mTOR inhibitors. *Drug Discovery Today*. 2011;16(7–8): 325–31. DOI: 10.1016/j.drudis.2011.02.008
- [144] Colombo N., McMeekin S., Schwartz P., Kostka J., Sessa C., Gehrig P., et al. A phase II trial of the mTOR inhibitor AP23573 as a single agent in advanced endometrial cancer. *Journal of Clinical Oncology*. 2007;25(18):5516.
- [145] Mackay H., Welch S., Tsao M.S., Biagi J.J., Elit L., Ghatge P., et al. Phase II study of oral ridaforolimus in patients with metastatic and/or locally advanced endometrial cancer; NCIC CTG IND 192. *Journal of Clinical Oncology*. 2011;29:s(5013).
- [146] Guertin D.A., Sabatini D.M. Defining the role of mTOR in cancer. *Cancer Cell*. 2007;12(1):9-22. DOI: <http://dx.doi.org/10.1016/j.ccr.2007.05.008>
- [147] Carracedo A., Pandolfi P.P. The PTEN-PI3K pathway: of feedbacks and cross-talks. *Oncogene*. 2008;27(41): 5527–5541. DOI: 10.1038/onc.2008.247
- [148] García-Martínez J.M., Moran J., Clarke R.G., Gray A., Cosulich S.C., Chresta C.M., et al. Ku-0063794 is a specific inhibitor of the mammalian target of rapamycin (mTOR). *Biochemical Journal*. 2009;421(1):29-42. DOI: 10.1042/BJ20090489
- [149] Feldman M.E., Apsel B., Uotila A., Loewith R., Knight Z.A., Ruggero D., et al. Active-site inhibitors of mTOR target rapamycin-resistant outputs of mTORC1 and mTORC2. *PLoS Biology*. 2009;7(2):e38. DOI: 10.1371/journal.pbio.1000038

- [150] 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 Research*. 2009;69(15):6232-6240. DOI: 10.1158/0008-5472.CAN-09-0299
- [151] Chresta C.M., Davies B.R., Hickson I., Harding T., Cosulich S., Critchlow S.E., et al. AZD8055 is a potent, selective, and orally bioavailable ATP-competitive mammalian target of rapamycin kinase inhibitor with in vitro and in vivo antitumor activity. *Cancer Research*. 2010;70(1):288-298. DOI: 10.1158/0008-5472.CAN-09-1751
- [152] Yu K., Shi C., Toral-Barza L., Lucas J., Shor B., Kim J.E., et al. Beyond rapalog therapy. Preclinical pharmacology and antitumor activity of WYE-125132, an ATP-competitive and -specific inhibitor of mTORC1 and mTORC2. *Cancer Research*. 2010;70(2):621-631. DOI: 10.1158/0008-5472.CAN-09-2340
- [153] Bhagwat V.S., Gokhale P.C., Crew A.P., Cooke A., Yao Y., Mantis C., et al. Preclinical characterization of OSI-027, a potent and selective inhibitor of mTORC1 and mTORC2: distinct from rapamycin. *Molecular Cancer Therapeutics*. 2011;10(8):1394-1406. DOI: 10.1158/1535-7163.MCT-10-1099
- [154] Thoreen C.C., Kang S.A., Chang J.W., Liu Q., Zhang J., Gao Y., et al. An ATP-competitive mammalian target of rapamycin inhibitor reveals rapamycin-insensitive functions of mTORC1. *Journal of Biological Chemistry*. 2009;284(12):8023-8032. DOI: 10.1074/jbc.M900301200
- [155] Zhang H., Berel D., Wang Y., Li P., Bhowmick N.A., Figlin R.A., et al. A comparison of Ku0063794, a dual mTORC1 and mTORC2 inhibitor, and temsirolimus in preclinical renal cell carcinoma models. *PLoS One*. 2013;8(1):e54918. DOI: 10.1371/journal.pone.0054918
- [156] Blaser B., Waselle L., Dormond-Meuwly A., Dufour M., Roulin D., Demartines N., et al. Antitumor activities of ATP-competitive inhibitors of mTOR in colon cancer cells. *BMC Cancer*. 2012;12:86. DOI: 10.1186/1471-2407-12-86
- [157] Liu Q., Wang J., Kang S.A., Thoreen C.C., Hur W., Ahmed T., et al. Discovery of 9-(6-aminopyridin-3-yl)-1-(3-(trifluoromethyl)phenyl)benzo[h][1,6]naphthyridin-2(1H)-one (Torin2) as a potent, selective, and orally available mammalian target of rapamycin (mTOR) inhibitor for treatment of cancer. *Journal of Medicinal Chemistry*. 2011;54(5):1473-80. DOI: 10.1021/jm101520v
- [158] Patricelli MP, Nomanbhoy TK, Wu J, Brown H, Zhou D, Zhang J, et al. In situ kinase profiling reveals functionally relevant properties of native kinases. *Chemistry and Biology*. 2011;18(6):699-710. DOI: 10.1016/j.chembiol.2011.04.011
- [159] Liu Q., Xu C., Kirubakaran S., Zhang X., Hur W., Liu Y., et al. Characterization of Torin2, an ATP-competitive inhibitor of mTOR, ATM and ATR. *Cancer Research*. 2013;73(8):2574-86. DOI: 10.1158/0008-5472

- [160] Rodrik-Outmezguine V., Chandarlapaty S., Pagano N., Poulikakos P.I., Scaltriti M., Moskatel E., et al. mTOR kinase inhibition causes feedback-dependent biphasic regulation of AKT signaling. *Cancer Discovery*. 2011;3:248-259. DOI: doi: 10.1158/2159-8290.CD-11-0085
- [161] Gungor H., Saleem A., Agarwal R., Blagden S.P, Michael A., Stronach M., et al. Pharmacokinetic (PK)/pharmacodynamic (PD) analysis of escalating repeat doses of the AKT inhibitor GSK2141795 (GSK795) in patients (pts) with ovarian cancer. *Journal of Clinical Oncology*. 2011;29(15):5064.
- [162] Cope C.L., Gilley R., Balmanno K., Sale M.J., Howarth K.D., Hampson M., et al. Adaptation to mTOR kinase inhibitors by amplification of eIF4E to maintain cap-dependent translation. *Journal of Cell Science*. 2014;127(4):788-800. DOI: 10.1242/jcs.137588
- [163] Ducker G.S., Atreya C.E., Simko J.P., Hom Y.K., Matli M.R., Benes CH., et al. Incomplete inhibition of phosphorylation of 4E-BP1 as a mechanism of primary resistance to ATP-competitive mTOR inhibitors. *Oncogene*. 2014;33(12):1590-1600. DOI: 10.1038/onc.2013.92
- [164] Maira S.M., Stauffer F., Brueggen J., Furet P., Schnell C., Fritsch C., et al. Identification and characterization of NVP-BEZ235, a new orally available dual phosphatidylinositol 3-kinase/mammalian target of rapamycin inhibitor with potent in vivo antitumor activity. *Molecular Cancer Therapy*. 2008;7(7):1851-1863. DOI: 10.1158/1535-7163.MCT-08-0017
- [165] Park S., Chapuis N., Bardet V., Tamburini J., Gallay N., Willems L., et al. PI-103, a dual inhibitor of Class IA phosphatidylinositol 3-kinase and mTOR, has antileukemic activity in AML. *Leukemia*. 2008;22(9):1698-1706. DOI: 10.1038/leu.2008.144
- [166] Mallon R., Hollander I., Feldberg L., Lucas J., Soloveva V., Venkatesan A., et al. Antitumor efficacy profile of PKI-402, a dual phosphatidylinositol 3-kinase/mammalian target of rapamycin inhibitor. *Molecular Cancer Therapeutics*. 2010;9(4):976-984. DOI: 10.1158/1535-7163.MCT-09-0954.
- [167] Wallin J.J., Edgar K.A., Guan J., Berry M., Prior W.W, Lee L., et al. GDC-0980 is a novel class I PI3K/mTOR kinase inhibitor with robust activity in cancer models driven by the PI3K pathway. *Molecular Cancer Therapeutics*. 2011;10(12):2426-36. DOI: 10.1158/1535-7163.MCT-11-0446
- [168] Cheng H., Li C., Bailey S., Baxi S.M., Goulet L., Guo L., et al. Discovery of the highly potent PI3K/mTOR dual inhibitor PF-04979064 through Structure-based drug design. *ACS Medicinal Chemistry Letters*. 2012;4(1):91-97. DOI: 10.1021/ml300309h
- [169] Markman B., Tabernero J., Krop I., Shapiro G.I., Siu L., Chen L.C., 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. *Annals of Oncology*. 2012;23(9):2399-408. DOI: 10.1093/annonc/mds011

- [170] Papadopoulos K.P., Tabernero J., Markman B., Patnaik A., Tolcher A.W., 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. *Clinical Cancer Research*. 2014;20(9):2445-56. DOI: 10.1158/1078-0432.CCR-13-2403
- [171] Courtney K.D., Corcoran R.B., Engelman J.A. The PI3K pathway as drug target in human cancer. *Journal of Clinical Oncology*. 2010;28(6):1075-1083. DOI: 10.1200/JCO.2009.25.3641
- [172] O'Reilly K.E., Rojo F., She Q.B., Solit D., Mills G.B., Smith D., et al. mTOR inhibition induces upstream receptor tyrosine kinase signaling and activates Akt. *Cancer Research*. 2006;66(3):1500-1508. DOI: 10.1158/0008-5472.CAN-05-2925
- [173] Liu T.J., Koul D., LaFortune T., Tiao N., Shen R.J., Maira S.M., et al. NVP-BEZ235, a novel dual phosphatidylinositol 3-kinase/mammalian target of rapamycin inhibitor, elicits multifaceted antitumor activities in human gliomas. *Molecular Cancer Therapy*. 2009;8(8):2204-2210. DOI: 10.1158/1535-7163
- [174] Santiskulvong C., Konecny G.E., Fekete M., Chen K.Y., Karam A., Mulholland D., et al. Dual targeting of phosphoinositide 3-kinase and mammalian target of rapamycin using NVP-BEZ235 as a novel therapeutic approach in human ovarian carcinoma. *Clinical Cancer Research*. 2011;17(8):2373-2384. DOI: 10.1158/1078-0432.CCR-10-2289
- [175] Sanchez C.G., Ma C.X., Crowder R.J., Guintoli T., Phommaly C., Gao F., et al. Preclinical modeling of combined phosphatidylinositol-3-kinase inhibition with endocrine therapy for estrogen receptor-positive breast cancer. *Breast Cancer Research*. 2011;13(2):1-17. DOI: 10.1186/bcr2833
- [176] O'Brien N.A., McDonald K., Tong L., von Euw E., Kalous O., Conklin D., et al. Targeting PI3K/mTOR overcomes resistance to HER2-targeted therapy independent of feedback activation of AKT. *Clinical Cancer Research*. 2014;20(13):3507-3520. DOI: 10.1158/1078-0432.CCR-13-2769
- [177] Schnell C.R., Stauffer F., Allegrini P.R., O'Reilly T., McSheehy P.M., Dartois C., et al. Effects of the dual phosphatidylinositol 3-kinase/mammalian target of rapamycin inhibitor NVP-BEZ235 on the tumor vasculature: implications for clinical imaging. *Cancer Research*. 2008;68(16):6598-6607. DOI: 10.1158/0008-5472.CAN-08-1044
- [178] Cao P., Maira S.M., Garcia-Echeverria C., Hedley D.W. Activity of a novel, dual PI3 kinase/mTor inhibitor NVP-BEZ235 against primary human pancreatic cancers grown as orthotopic xenografts. *British Journal of Cancer*. 2009;100(8):1267-1276. DOI: 10.1038/sj.bjc.6604995
- [179] Marone R., Erhart D., Mertz A.C., Bohnacker T., Schnell C., Cmiljanovic V., et al. Targeting melanoma with dual phosphoinositide 3-kinase/mammalian target of rapamycin inhibitors. *Molecular Cancer Research*. 2009;7(4):601-613. DOI: 10.1158/1541-7786.MCR-08-0366

- [180] McMillin D.W., Ooi M., Delmore J., Negri J., Hayden P., Mitsiades N., et al. Antimyeloma activity of the orally bioavailable dual phosphatidylinositol 3- kinase/mammalian target of rapamycin inhibitor NVP-BEZ235. *Cancer Research*. 2009;69(14):5835-5842. DOI: 10.1158/0008-5472.CAN-08-4285
- [181] Cho D.C., Cohen M.B, Panka D.J, Collins M., Ghebremichael M., Atkins M.B., et al. The efficacy of the novel dual PI3- kinase/mTOR inhibitor NVP-BEZ235 compared with rapamycin in renal cell carcinoma. *Clinical Cancer Research*. 2010;16(14):3628–3638. DOI: 10.1158/1078-0432.CCR-09-3022
- [182] Cloughesy T.F., Yoshimoto K., Nghiemphu P., Brown K., Dang J., Zhu S., et al. Anti-tumor activity of rapamycin in a Phase I trial for patients with recurrent PTEN-deficient glioblastoma. *PLoS Medicine*. 2008;5(1):e8. DOI: 10.1371/journal.pmed.0050008
- [183] 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. *British Journal of Cancer*. 2012;107(7):1093-9. DOI: 10.1038/bjc.2012.368
- [184] Asahina H., Nokihara H., Yamamoto N., Yamada Y., Tamura Y., Honda K., et al. Safety and tolerability of AZD8055 in Japanese patients with advanced solid tumors; a dose-finding phase I study. *Investigational New Drugs*. 2013;31(3):677-84. DOI: 10.1007/s10637-012-9860-4
- [185] Tan D.S., Dumez H., Olmos D., Sandhu S.K., Hoeben A., Stephens A.W. First-in-human phase I study exploring three schedules of OSI-027, a novel small molecule TORC1/TORC2 inhibitor, in patients with advanced solid tumors and lymphoma. *Journal of Clinical Oncology*. 2010;28:(15s).Abstract: 3006

