



# The PI3K/AKT/mTOR signaling pathway inhibitors enhance radiosensitivity in cancer cell lines

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## Abstract

**Introduction** Radiotherapy is one of the most common types of cancer treatment modalities. Radiation can affect both cancer and normal tissues, which limits the whole delivered dose. It is well documented that radiation activates phosphatidylinositol 3-kinase (PI3K) and AKT signaling pathway; hence, the inhibition of this pathway enhances the radiosensitivity of tumor cells. The mammalian target of rapamycin (mTOR) is a regulator that is involved in autophagy, cell growth, proliferation, and survival.

**Conclusion** The inhibition of mTOR as a downstream mediator of the PI3K/AKT signaling pathway represents a vital option for more effective cancer treatments. The combination of PI3K/AKT/mTOR inhibitors with radiation can increase the radiosensitivity of malignant cells to radiation by autophagy activation. Therefore, this review aims to discuss the impact of such inhibitors on the cell response to radiation.

**Keywords** PI3K/AKT · mTOR · Radiation · Inhibitor · Radiosensitivity

## Introduction

PI3K/AKT/mTOR signaling pathway participates in a wide range of cell biological processes such as cell growth, cell survival, etc. In the PI3K/AKT/mTOR signaling pathway, PI3K stands for phosphatidylinositide 3-kinases (s), AKT for a serine/threonine kinase or protein kinase B, and mTOR for mammalian target of rapamycin. The majority of cancers show hyperactivation of this pathway [1]. mTOR, a 289-kDa serine-threonine protein kinase, acts as a central regulator of cell proliferation, growth, metabolism, survival, and autophagy [2]. The signaling pathway of this mediator gets activated in a variety of cellular physiological processes including tumorigenesis, angiogenesis, insulin resistance,

and T cells activation [3]. Besides, mTOR as a downstream kinase of phosphatidylinositol 3-kinase/AKT (PI3K/AKT) signaling pathway is closely paired with neoplastic disorders [4]. This pathway is a key cascade downstream of different protein kinases such as epidermal growth factor receptor (EGFR) family members (Fig. 1) [5]. It is well known that mTOR plays a major role in the inactivation of the translation suppressor eukaryotic initiation factor 4E-binding protein 1 (4E-BP1, PHAS1) and activates the ribosomal p70S6 kinase through phosphorylation [6]. To that end, mTOR is a promising option for the clinical development of effective molecular targeted therapies. Disruption of mTOR and other protein molecules in this signaling pathway often takes place in various human cancers and type 2 diabetes [7]. Moreover, tumor cells are more susceptible to mTOR inhibitors in comparison with normal cells [8]. Different reports suggested the correlation of mTOR signaling pathway hyperactivity with colorectal cancers [9], adenocarcinomas [10], glioblastomas [11], and astrocytomas [12], which leads to their higher sensitivity to mTOR inhibitors.

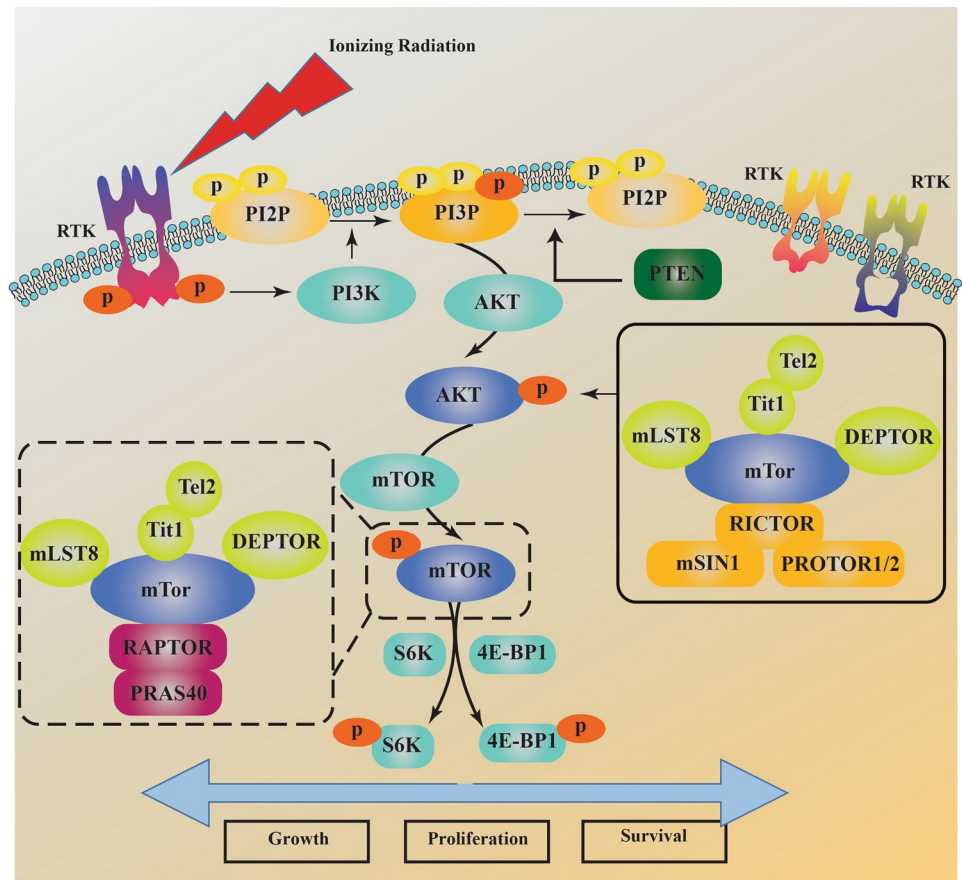
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**Fig. 1** The PI3K/AKT/mTOR signaling pathway: ligand binding or ionizing radiation triggers the activation of PI3K, which leads to the phosphorylation of phosphatidylinositol-4,5-bisphosphate (PIP<sub>2</sub>) and production of phosphatidylinositol-3,4,5-trisphosphate (PIP<sub>3</sub>). PTEN antagonizes the kinase activity of PI3K by dephosphorylation of PIP<sub>3</sub>. As a phosphate source PIP<sub>3</sub> transfers a phosphate group to AKT and activates it. AKT has different regulatory sites that can be phosphorylated by the mTORC2 complex. AKT as a kinase phosphorylates different downstream targets such as mTORC1 which eventually results in the cell growth, survival, and proliferation



## PI3K/AKT/mTOR signaling pathway

PI3Ks, as a family of intracellular lipid kinases, phosphorylate the 3'-OH functional group of phosphatidylinositides (PIs). Three different classes of PI3Ks are discovered. The most prominent one is class 1, which converts membrane-bound PI-(4,5)-bisphosphate [PI(4,5)P<sub>2</sub>; PIP<sub>2</sub>] to PI-(3,4,5)-trisphosphate [PI(3,4,5)P<sub>3</sub>; PIP<sub>3</sub>]. PIP<sub>3</sub> is considered a secondary messenger that triggers the recruitment and activation of kinases mainly those that possess pleckstrin homology (PH) domain, as an example PI3K-dependent kinase-1 (PDK1) [13]. Phosphatase and tensin homolog (PTEN) as a naturally occurring negative regulator of PI3K/AKT/mTOR pathway effectively suppresses tumor (Fig. 1). PTEN can dephosphorylate PIP<sub>3</sub>; hence, it determines the signaling duration of PIP<sub>3</sub>. PTEN shows mutation or deletion in a wide range of human cancers; and in some cancers, it is epigenetically silenced [14]. The downstream kinase of PI3K is AKT [a serine/threonine kinase or protein kinase B (PKB)]. The complete activation of AKT happens when PDK1 and mTORC2 phosphorylate its T308 and S473 residues. The activated AKT phosphorylates a wide range of other enzymes including glycogen synthase

kinase 3 (GSK3), tuberous sclerosis 2 (TSC2), caspase 9, and PRAS40 (AKT1S1) [15]. To that end, AKT is involved in a vast range of cellular activities such as cell proliferation, differentiation, apoptosis, angiogenesis, and metabolism. mTOR is a downstream kinase of PI3K/AKT which belongs to the PI3K-related kinase family (PIKK). This mediator is involved in at least two structurally different multi-protein complexes namely mTORC1 and mTORC2 [16, 17]. mTORC1 consists of mTOR (the catalytic subunit), Raptor (regulatory-associated protein of mTOR), mLST8 (mammalian lethal with Sec13 protein 8 also known as GβL), Deptor (DEP-domain-containing mTOR-interacting protein), and PRAS40 (proline-rich AKT substrate 40 kDa) [16, 18]. The mTORC2 includes mTOR, mLST8, Deptor, Rictor (rapamycin-insensitive companion of mTOR), mSIN1 (mammalian stress-activated protein kinase interacting protein 1 or MAPKAP1), and Protor-1 (protein observed with Rictor-1) [17]. mTORC1 can be regulated by various environmental signals such as hormones, growth factors, nutrients, and cellular stress, in which other mediators such as RHEB (RAS homolog enriched in brain), guanosine triphosphatases (GTPase), and TSC1/2 display a crucial role [19]. Moreover, the PI3K/AKT signaling pathway is a critical upstream regulator of mTORC1 [20]. mTORC1 is a rapamycin and

nutrient-sensitive complex which can influence the cell-growth and survival by increasing the biosynthesis of protein. Protein synthesis can be regulated by mTORC1 through phosphorylation of S6K1 and 4E-BP1 (Fig. 1) [21]. mTORC2 is also called the rapamycin-insensitive complex and can be involved in the regulation of the cytoskeletal organization. It can also play a role in the regulation of cell survival and proliferation by phosphorylation and activation of AKT while the exact downstream of mTORC2 is unclear [22]. It is worth noting that Tti1 is another positive regulatory protein for the mTOR complexes. Tti1 interacts with telomere maintenance 2 (Tt2) and regulates the stability of phosphatidylinositol 3-kinase-related protein kinase family members (Fig. 1) [23].

### **Ionizing radiation, radioresistance, and PI3K/AKT/mTOR signaling pathway**

Radiation resistance is a fundamental problem in radiotherapy. Understanding the mechanisms and signaling pathways of radioresistance plays a vital role in the development of strategies to overcome tumor radioresistance in cancer therapy. These strategies include at least two different approaches such as radiotherapy and chemotherapy (for instance mTOR signaling pathway inhibitors). The rapid development in biomedical science through advanced techniques like genomics and proteomics caused a huge amount of attention toward radioresistance related signaling pathways. The theory of radiation therapy is that DNA damages [such as single-strand breaks (SSBs) and double-strand breaks (DSBs)] result in apoptosis and subsequently tumor regression [24]. However, activation of some major pathways such as PI3K/AKT/mTOR signaling pathway can increase tumor cell survival even after radiation and jeopardizes the efficacy of the therapy procedure. In the presence of radiation, this signaling pathway induces tumor cell proliferation, growth, and survival. Different studies suggest that PI3K/AKT pathway controls the cell cycle progression and cell proliferation through translation of specific mRNA transcripts taking advantage of its downstream mediator mTOR. For instance, mTOR increase the expression of receptor tyrosine kinases such as EGFR (Fig. 1) [25, 26].

### **Strategies to inhibit PI3K/AKT/mTOR signaling pathway**

A wide range of small molecules has been introduced as PI3K/AKT/mTOR signaling pathway inhibitors. These compounds fall into two major groups including single inhibitors that only inhibit PI3K, AKT, or mTOR signaling mediators and dual inhibitors that simultaneously inhibit PI3K and mTOR signaling proteins. The inhibition of PI3K/AKT/mTOR signaling pathway mediators make cancer cells more

susceptible to radiotherapy, anti-EGFR treatment, and other receptor tyrosine kinase (RTK) inhibitor therapies.

### **ATP-competitive mTOR kinase inhibitors**

A new Group of mTOR inhibitors, that bind to the ATP binding region of mTOR and suppress the kinase-dependent signaling activities of mTOR complexes, are known as ATP-competitive mTOR kinase inhibitors (TKIs). The advantage of these compounds is that they not only block the kinase activity of both mTORC1 and mTORC2, but also inhibits the feedback activation of the PI3K/AKT pathway [27]. These inhibitors include WYE-354 [28], WYE-687 [29], PP242 [30], Torin 1 [31], and KU-0063794 [32] which display a potent inhibition of mTOR pathway. Like rapamycin derivatives, TKIs can decrease protein translation, attenuate cell proliferation, and block angiogenesis in several cancer cell lines. In fact, cell proliferation inhibition by TKIs inhibitors is more potent than rapalogs [33].

### **Dual mTOR/ PI3K inhibitors**

The interaction of mTOR with the PI3K signaling network triggers the development of dual mTOR/ PI3K inhibitors. These inhibitors target the PI3K $\alpha$ ,  $\beta$ , and  $\gamma$  isoforms as well as the ATP-binding sites of both mTORC1/mTORC2; hence, they suppress the PI3K/AKT/mTOR signaling pathway. Blocking the biological function of PI3K/mTOR signaling pathway components has been shown to inhibit tumor cell proliferation. Apoptosis and autophagy induction has also been observed in cell treatment with these compounds [34].

### **Rapamycin and its analogs (rapalogs)**

Rapamycin, a potent mTORC1 inhibitor, leads to the inhibition of cell growth, cell proliferation, and cell cycle progression. However, due to the slight aqueous solubility and stability, its application is limited for cancer treatments [35]. Therefore, different rapamycin analogs (rapalogs) with more desirable pharmaceutical properties have been developed, such as CCI-779 (Temsirrolimus), RAD001 (Everolimus), AP23573 (Deforolimus), SAR943 (32-deoxorapamycin) or ABT-578 (Zotarolimus) for cancers [36, 37], allergic inflammation [38, 39] or coronary stent implantation [40]. Pre-clinical studies confirmed their antiproliferative effect on several types of cancer. Furthermore, clinical studies have revealed favorable anticancer effectiveness in certain cancers [37, 41]. The mechanism of rapamycin action is not completely understood; however, it is admitted that rapamycin and its analog strongly bind to the FKBP12-rapamycin binding (FRB) domain and inhibits the mTORC1 assembly and its kinase activity [42]. In contrast to mTORC1, rapamycin and its analog have a much lower affinity toward mTORC2;

however, prolonged drug exposure has been demonstrated to inhibit mTORC2 formation. Unlike mTORC1 inhibition, prolonged rapamycin treatment indirectly inhibits mTORC2. Rapalogs finally exert their impacts by causing changes in multiple downstream pathways including cell proliferation and angiogenesis. A possible mechanism for the antiproliferative impacts is the prevention of 4E-BP1 and S6K1 phosphorylation by mTORC1 [43].

### The combination of mTOR inhibitors and radiation

mTOR inhibitors combined with radiotherapy can be favorable agents for cancer therapy [44, 45].

#### Rapamycin

Rapamycin is a macrolide compound that binds to FKBP-12 (FK506 binding protein 12 which plays a vital role in immunoregulation, essential cellular processes including membrane protein folding and cellular trafficking). Rapamycin, the allosteric inhibitor of mTOR, prevents major phosphorylation of proteins that are involved in the process of transcription, translation, and cell cycle regulation [46]. A great number of cancer cells with mutations in the PI3K/AKT-mTOR pathway are susceptible to the cell growth blocking induced by rapamycin. Rapamycin inhibited tumor growth in glioma xenografts treated with fractionated stereotactic radiotherapy. The local control of tumor cells can also be improved via the anti-angiogenesis effects induced by rapamycin during radiation therapy. Rapamycin significantly increased the effectiveness of fractionated radiation therapy in U87 (glioma cell line) xenograft models [47]. Dumont et al. have demonstrated that rapamycin combined with  $^{177}\text{Lu}$ -RM2 (37 MBq, RM2 is a DOTA-conjugated gastrin-releasing peptide receptor antagonist) resulted in prolonged survival in comparison with single agents (Table 1) [48]. A combination of rapamycin treatment and  $^{137}\text{Cs}$  (6.4 Gy/min) was investigated in a glioblastoma multiforme (GBM) model using U87 xenograft models. The treatment with low-intensity radiation (16 Gy/18 days) did not have considerable effects on tumor growth rates (regrowth delay of  $-0.2 \pm 4.6$  days) compared with control in the xenograft models. However, rapamycin in combination with radiation therapy considerably improved the delayed regrowth ( $19.1 \pm 6.3$  days) in comparison with rapamycin alone; Hence, combination treatment with rapamycin and radiation was noticeably more effective than both radiation alone and rapamycin [49]. Rapamycin at different concentrations (1–8 nM) plus radiation (dose rate of 300 cGy/min) was studied in glioblastoma cells. This study suggests that the pre-treatment of U87, T98, and A172 glioblastoma cells with rapamycin decreased survivin expression and clonogenic capacity which leads to more radiosensitivity of these

cell lines to radiotherapy (Table 1) [50]. A phase I/II clinical study on rectal cancer patients evaluated the feasibility of short-course radiotherapy (SCRT) in conjugation with rapamycin. Phase I of this study in 13 patients determined that the maximum tolerated dose (MTD) of rapamycin is 6 mg. Phase II of the clinical study evaluated the combination of rapamycin (6 mg) and radiotherapy in a population of 31 rectal cancer patients. This study recommends that SCRT and rapamycin combination is feasible and safe and resulted in tumor size shrinkage, while due to the small sample size no significant complete pathological response was observed. However, positron emission tomography (PET) scans following phase II treatment confirmed a significant ( $p < 0.05$ ) reduction in tumor size (Table 2) [51].

#### Temsirolimus

Temsirolimus (also called CCI-779) is the first FDA-approved mTOR inhibitor for advanced renal cell cancer treatment. Meanwhile, its clinical development also includes research studies in different tumors [52]. Ekshyyan et al. investigated the temsirolimus capability to radio-sensitize head and neck squamous cell carcinoma (HNSCC) in both in vitro and in vivo models [53]. Temsirolimus combined with radiation therapy was far more impressive in tumor cell growth inhibition in comparison with cisplatin plus radiation in murine xenografts. Studies displayed that radiation in the presence of temsirolimus sharply reduced radiation-induced activation of the mTOR signaling pathway, enhanced apoptosis, and decreased the number of blood vessels in tumors [54]. The radiosensitivity of A549 cells was studied under both normoxia and hypoxia conditions by using temsirolimus. A549 cells revealed a radio-resistance of 5.1 Gy under normoxia and 14.2 Gy under hypoxia, as the D10 values, the oxygen enhancement ratio (OER) was also 2.8. The A549 cell line survival rate with temsirolimus under hypoxia conditions significantly decreased in comparison with those in normoxia. The D10 values were 4.8 and 5.4 Gy in normoxia and hypoxia conditions, respectively (OER = 1.1). Temsirolimus suppressed the expression of mTOR in both normoxia and hypoxia. The HIF-1 $\alpha$  expression also decreased under hypoxia by temsirolimus. Therefore, temsirolimus could overcome the cancer radio-resistance caused by hypoxia [55].

#### Everolimus

Everolimus, also known as RAD001, is the 40-O-(2-hydroxyethyl) analog of rapamycin that acts similarly to rapamycin as a mTOR inhibitor. Cao et al. studied the capability of everolimus to improve radiation therapy effects on two CaP (prostate cancer) cell lines, PC-3 and DU145, and demonstrated that both cell types became more sensitive

to radiation after everolimus therapy; however, PC-3 cells showed higher radiation sensitivity [56]. YU et al. evaluated the effects of everolimus treatment after irradiating the SCC4 cells with ionizing radiation. RAD001 improved the radio-sensitivity of SCC4 cells treated using 6 Gy radiation without any effect on cell death. Everolimus inhibited the mTOR, and S6, as well as 4E-BP1 phosphorylation and reduced the tumor cells' clonogenic survival after irradiation. Moreover, the combination of everolimus (300 nM) and 6 Gy radiation considerably suppressed SCC4 cell growth in comparison with both 6 Gy radiation alone and everolimus (30 nM) combined with 6 Gy radiation [57]. The effect of the combination of Z-DEVD (caspase-3 inhibitor), everolimus, and  $^{137}\text{Cs}$  (1.8 Gy/min) was studied on H460 (a human large-cell lung carcinoma line) cell line. In comparison with radiation alone, the combination treatment substantially delayed tumor growth (32 vs. 23 days). The combination treatment resulted in a sharp reduction of the proliferation index (by reducing proliferative marker Ki67 levels) compared with radiation therapy alone ( $13 \pm 2$  vs.  $38 \pm 1.5$ ). These findings revealed that a combination of Z-DEVD and everolimus can enhance the radiosensitivity in H460 cells (Table 1) [58].

The effect of everolimus in combination with radiation and fluorouracil (5-FU) on rectal cancer patients was evaluated during the phase I/II clinical trial. The MTD for everolimus was 10 mg. the patients were treated with everolimus 14 days before and during chemoradiotherapy (four weeks). No significant complete pathological response was observed (Table 2) [59].

Everolimus in combination with radiation was studied during a phase-I clinical trial for the treatment of locally advanced prostate cancer. The study determined that MTD of everolimus is 7.5 mg/day, while due to the minor side effects (Dose-limiting toxicity occurred in two out of fifteen patients) the administration of a smaller dose (5 mg/day) was recommended [60]. In parallel with this study other reports also mentioned that everolimus is well tolerable below 10 mg/day (Table 2) [61].

A clinical trial phase-I investigated the combination of everolimus, cisplatin, and pelvic radiotherapy for the treatment of locally advanced cervix cancer. In this study patients received everolimus 7 days prior and during the radiotherapy or brachytherapy (2.5, 5, and 10 mg/day) and the MTD was determined to be 5 mg/day (Table 2) [62].

A trial phase-I and two clinical trials phase-II investigated the administration of everolimus for the treatment of glioblastoma multiform (GBM). All these studies applied a treatment regimen composed of everolimus, radiotherapy and, temozolomide. Clinical trial phase-I indicated that MTD of everolimus is 10 mg/day (70 mg/week) (Table 2) [63]. A phase-II clinical trial indicated that everolimus does not improve the efficacy of standard chemoradiotherapy with radiotherapy/temozolomide. Moreover, the study indicated

the administration of 70 mg/week (MTD = 10 mg/day) of everolimus leads to moderate toxicity [64]. Another similar clinical trial phase-II indicated that the addition of everolimus (10 mg/day, 70 mg) to standard chemoradiotherapy does not improve progression-free survival (Table 2) [65].

The feasibility of everolimus combination with thoracic radiotherapy followed by consolidation chemotherapy in untreated non-small-cell lung cancer (NSCLC) was also investigated. The patients were treated with 50 mg/week of everolimus (according to other studies). The treatment appears to be safe and feasible while the authors recommend close observation for pulmonary toxicity [66].

### **Voxtalisib (SAR245409, XL765)**

Voxtalisib is a dual PI3K/mTOR inhibitor (Fig. 2). The effect of voxtalisib has been evaluated against a panel of glioblastoma multiform (GBM) cell lines and xenografts in combination with temozolomide [67]. A phase-I clinical trial evaluated the effect of voxtalisib on patients with high-grade glioblastoma in combination with temozolomide, with or without irradiation. The administration of voxtalisib was evaluated to be safe with MTD of 90 mg/day and moderate inhibition of PI3K/mTOR signaling pathway (Table 2) [68].

### **Dactolisib (NVP-BE235)**

Dactolisib, an imidazo[4,5-c]quinoline analog, is a dual PI3K/mTOR inhibitor and inhibits radiation-induced DSBs repair by inhibition of DNA-dependent protein kinase catalytic subunit (DNA-PKcs) and ataxia-telangiectasia mutated (ATM) in GBM cell lines [69]. Cerniglia et al. studied the effect of dactolisib in head and neck cancer cells (Fig. 2) [70]. Dactolisib impressively increased radio-sensitivity in HNSCC cells and weakened the DNA repair processes by decreasing the DNA-dependent protein kinase, catalytic subunit (DNA-PKCS) phosphorylation. Dactolisib therapy induces autophagy and the combination of this compound with radiation in the presence of autophagy inhibitors enhanced cell death (Table 1) [71].

The studies also suggest that dactolisib sensitize colorectal cancer (CRC) cell lines (HCT116, SW 620, and HT29) to radiation. In the presence of 5 Gy radiation (up to three hours), the phosphorylated AKT and mTOR were gradually increased which resulted in radioresistance. The combination of radiation and dactolisib increased the radiosensitivity of all the CRC cell lines. The study suggests that dactolisib reduces cell proliferation by lowering the phosphorylation of DNA-dependent protein kinases (DNA-PKs) and Ribosomal Protein S6 (rpS6) (a protein involved in cell growth and proliferation) by inhibiting the AKT and mTOR phosphorylation. A significant ( $p$ -value  $< 0.001$ ) reduction in tumor size was observed when an HCT116 xenograft received a

**Table 1** Preclinical studies investigating the effect of combined PI3K/AKT/mTOR inhibitors and radiation on different cell lines

Inhibitor	Radiation	Cell line	The observed effect	References
Rapamycin	<sup>177</sup> Lu-RM2	PC-3	Prolonged survival	[48]
	<sup>137</sup> Cs	U87	Delayed regrowth	[49]
	X-ray	U87, T98, and A172	Increased $\gamma$ -H2AX expression, Decreased surviving expression, and clonogenic capacity	[50]
Temsirrolimus	X-ray	HNSCC	Enhanced apoptosis, Decreased the blood vessel number in the tumor	[54]
		A549	Suppressed expression of mTOR, Decreased HIF-1 $\alpha$ expression	[55]
Everolimus	<sup>137</sup> Cs	PC-3 and DU145	Higher radiation sensitivity	[56]
	X-ray	SCC4	Decreased clonogenic survival, Suppressed cell growth	[57]
	<sup>137</sup> Cs	H460	Delayed tumor growth, Enhanced radio-sensitivity	[58]
Dactolisib	X-ray	HNSCC	DNA repair inhibition, Enhanced cell death	[71]
		HCT116, SW 620, and HT29	A combination of dactolisib and radiation reduced the tumor size of an HCT116 xenograft. Dactolisib enhanced apoptosis and inhibits DSB repair pathways	[72, 73]
		PC-3, DU145 and, LNCaP	Suppressing colony formation, increasing apoptosis, cell cycle arrest at G2/M phase, increased DSB of DNA, reduced inactivation of cell cycle checkpoint and DNA repair	[74]
Gedatolisib		UMSCC1-wtP53 and UMSCC46-mtP53	Delays tumor growth of UMSCC1 xenograft	[80]
		UT-SCC-14 and UT-SCC-15	Has no additive effect neither in vitro nor in vivo when combined with irradiation	[81]
Vistusertib		GSCs	Enhanced survival, DNA repair inhibition	[82]
Sapanisertib		PSN1, Panc1, and Miapaca-2	DNA repair inhibition, Cap complex formation	[83]
Torkinib	<sup>64</sup> Cu	IBC	High tumor control, Improved radiosensitivity	[84]
Torin2	X-ray	MCF 7	DNA repair inhibition, Growth inhibition	[85]
AZD8055		NPC	Cell proliferation and clonogenicity inhibition, delayed tumor growth, more apoptosis	[86]
SF1126		SF767, LNpuro, and LNVIII	Tumor growth inhibition, improved radio-sensitivity	[87]
3-Methyladenine		EC9706	Autophagy inhibition, enhanced cell apoptosis, and reduced tumor proliferation	[88]
LY294002		U251	DNA repair suppression, tumor cell death, high toxicity	[90, 91]
PI-103		HCT116, DLD1	Delayed G2/M phase, improved radio-sensitivity	[95]
		SW480, SW48	In combination with NVP-AUY922 (Heat Shock Protein 90 (HSP-90) inhibitor) effectively increases the radio-sensitivity	[96]
Omipalisib		NPC	DNA damage enhancement delayed G2/M phases and enhanced cell apoptosis	[97]
Palomid-529	X-ray	NCI-60 cell lines panel	Reduced the pAKT level in a large variety of cell lines. The combination of palomid-529 (20 mg/Kg) and radiation (6 Gy) led to 77.4% tumor shrinkage in PC-3 xenograft	[99]
HS-173		Miapaca-2 and PANC-1	Radiation and HS-173 induced a greater degree of G2/M arrest (91%) than radiation alone (75%). HS-173 improves radiosensitivity of human pancreatic cancer cells by inhibiting radiation induced DNA damage repair	[100]

combination of radiation and dactolisib [72]. Another similar study on the same CRC cell lines and xenograft indicated that prolonged administration of dactolisib after radiotherapy leads to enhanced apoptosis and inhibits DSB repair pathways (Table 1) [73].

Other reports suggest dactolisib improves radiosensitization of radioresistant prostate cancer (CaP) cell lines including (PC-3, DU145 and, LNCaP). This study concluded that dactolisib as a dual PI3K/mTOR inhibitor in combination with radiation is more effective against CaP cell lines in

**Table 2** Clinical trials of combined PI3K/AKT/mTOR inhibitors and radiation on different patients

Inhibitor	Cancer (no. of patients)	Stage of disease	Clinical trial phase	Radiotherapy dose (fraction no. × radiation)	Results		References
					MTD*	Clinical observations	
Rapamycin	Rectal cancer (phase-I (n = 13), phase-II (n = 31))	Primary resectable rectal cancer	Phase-I/II	Short-course hypofractionated radiotherapy (5 × 5 Gy)	6 mg	Phase II of the clinical study evaluated the combination of rapamycin and radiotherapy in a population of 31 rectal cancer patients. Significant reduction in tumor metabolic activity but no significant complete pathological response was observed	[51]
Everolimus	Rectal cancer (n = 12)	Primary resectable rectal cancer	Phase-I/II	(28 × 1.8 Gy)/day, 5 days/week	10 mg	Everolimus was administered in combination with radiation and fluorouracil (5-FU) and no significant complete pathological response or increment in toxicity was observed. Further evaluation was suggested	[59]
	Prostate cancer (n = 14)	Primary locally advanced non-metastatic	Phase-I	37 × 2 Gy	5–7.5 mg	Due to the minimal side effects, the recommended MTD is 5 mg/day	[60]
	Prostate cancer (n = 18)	Biochemical recurrence after the prostatectomy	Phase-I	37 × 1.8 Gy	≤ 10 mg	No dose-limiting toxicities were observed	[61]
	Cervix cancer (n = 13)	IIB, IIIA or IIIB	Phase-I	25 × 1.8 Gy for radiotherapy and 4 × 6 Gy for brachytherapy	5 mg	The combination of everolimus, cisplatin, and pelvic radiotherapy for locally advanced cervix cancer is safe and needs further studies	[62]
	GBM (n = 18)	Just diagnosed	Phase-I	30 × 2 Gy	10 mg	Everolimus in combination with irradiation and adjuvant temozolomide was reasonably tolerable	[63]
	GBM (n = 100)	Just diagnosed	Phase-II	30 × 2 Gy	10 mg	The addition of everolimus to standard irradiation/temozolomide chemoradiotherapy has no significant impact on the treatment and caused moderate toxicity	[64]
	GBM (n = 171)	Just diagnosed	Phase-II	30 × 2 Gy	10 mg	A combination of everolimus and standard irradiation/temozolomide was applied. The increased treatment-related toxicities with no improvement in the patients' progression-free survival were observed	[65]

Table 2 (continued)

Inhibitor	Cancer (no. of patients)	Stage of disease	Clinical trial phase	Radiotherapy dose (fraction no. X radiation)	Results		References
					MTD*	Clinical observations	
	NSCLC (n = 26)	Stage III-IV	Phase-I	28–66 Gy in 14–33 fraction	–	Treatment is safe and practical however the observation for pulmonary toxicity is necessary	[66]
Voxtalisisb	GBM (n = 54)	Just diagnosed	Phase-I	1.8–2 Gy fractions up to 60 Gy, 5 days/week	90 mg	The administration of voxtalisisb was with moderate inhibition of PI3K/mTOR signaling pathway	[68]

\*Maximum tolerated dose

comparison with single inhibitors of PI3K/mTOR signaling pathway such as BKM120 and Rapamycin. After treatment with dactolisib, all the mentioned cell lines displayed lower colony formation, higher and cell cycle attest at G2/M phase. Moreover, DSB of DNA was increased and it reduced the expression level of proteins responsible for inactivation of cell cycle checkpoint and DNA repair [including non-homologous end joining (NHEJ) and homologous recombination (HR)] [74]. Other studies also confirmed that dactolisib causes cell cycle arrest at the G2/M phase and improves the radiosensitivity of PC3 and DU145 cell lines to radiation which is independent of oxygen concentration or PTEN mutation status (Table 1) [75, 76].

Breast cancer cell line (MDA-MB-231 and, MCF-7) displayed an improved radiosensitivity after treatment with dactolisib. This compound is a dual PI3K/mTOR inhibitor; hence, the observed radiosensitivity was independent of oxygen condition (hypoxia, normoxia and, reoxygenation) [77]. Wang et al. reported that dactolisib in combination with radiation increases autophagy, apoptosis, and cell cycle attest of glioma stem cells (SU-2 cell line) [78]. It is noteworthy, based on the drug and ionizing radiation schedule the observed effect of NVP-BEZ235 can vary from a strong radiosensitizer to a cytostatic agent in glioblastoma cells (including U87-MG, GaMG, and DK-MG) [79].

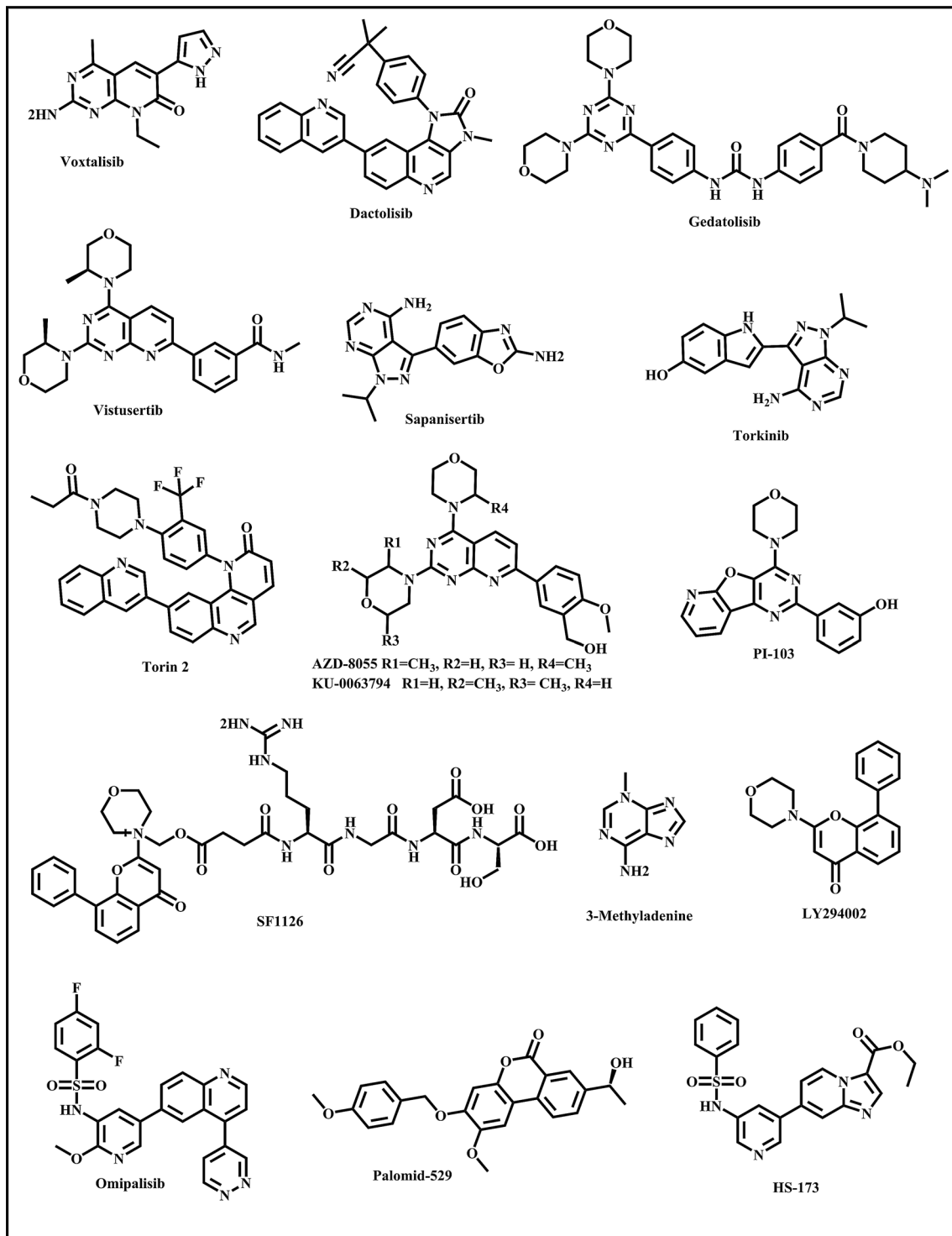
### Gedatolisib (PF-05212384, PKI-587)

Gedatolisib is a dual PI3K/MTOR inhibitor (Fig. 2). A study claimed that gedatolisib significantly radiosensitizes head and neck squamous cell carcinoma cell lines (UMSCC1-wtP53 and UMSCC46-mtP53) and delays tumor growth of UMSCC1 xenograft in combination with fractionated radiation [80]. Another study on different head and neck squamous cell carcinoma cell lines (UT-SCC-14 and UT-SCC-15) reported that gedatolisib has no additive effect neither in vitro nor in vivo when combined with radiotherapy. However, gedatolisib can significantly reduce the phosphorylation of key components in the PI3K/mTOR signaling pathway (Table 1) [81].

### Vistusertib (AZD2014)

Vistusertib is a selective inhibitor of mTOR with IC<sub>50</sub> of 2.8 nM (Fig. 2). Kahn et al. determined the effects of AZD2014 on the radiosensitivity of glioblastoma stem-like cells (GSCs). They found that AZD2014 plus X-ray (dose rate of 2.3 Gy/min) resulted in the inhibition of mTORC1 and mTORC2 complexes. The combination of AZD2014 and radiation considerably enhanced survival in comparison with AZD2014 and radiation treatment alone. AZD2014 improved the radiosensitivity of GSC cells (CD133 and





**Fig. 2** The chemical structure of PI3K/AKT/mTOR signaling pathway inhibitors

CD15 positive cells) by DNA repair inhibition (Table 1) [82].

### Sapanisertib (INK128)

Sapanisertib is an experimental mTORC1 and mTORC2 inhibitor with  $IC_{50}$  of 1 nM (Fig. 2). Hayman et al. studied the effects of sapanisertib on the radiosensitivity of pancreatic carcinoma cells. Sapanisertib inhibited the activity of both mTOR1 and mTOR2 in 3 pancreatic tumor cells (PSN1, Panc1, and Miapaca-2). Sapanisertib inhibited the DNA repair and formation of the cap complex in cancer cells. Sapanisertib in combination with radiation caused an enhancement of tumor radiosensitivity (Table 1) [83].

### Torkinib (pp242)

Torkinib is a potent and selective mTORC1 and mTORC2 ATP-competitive inhibitor with  $IC_{50}$  of 8 nM (Fig. 2). Connolly et al. evaluated the effects of mTOR inhibitors (pp242 and RAD001) in combination with radiation in IBC xenografts. It is suggested that pp242 significantly inhibited mTOR1 and mTOR2 activity in IBC cells and blocked the upregulation of both AKT and ERK. However, RAD001 inhibited mTORC1 activity and led to AKT activation. Torkinib plus radiation showed a high degree of tumor control and improved radiosensitivity in IBC cells in vitro, in comparison with RAD001 (Table 1) [84].

### Torin 2

Torin 2 is a selective mTOR ATP-competitive inhibitor (Fig. 2). Jia Luo et al. studied the effect of Torin2 on the radio-sensitivity of MCF7 cells (breast cancer cell line). They found that the activity of mTOR was repressed in MCF-7 cells treated with 20 nM Torin 2 before irradiation with X-rays at (0–10 Gy  $\leq$  1 min). Torin 2 pretreatment reduced the radiation-induced AKT/mTOR pathway activation. Moreover, Torin 2 partially blocked the DNA repair by reducing the ataxia telangiectasia mutated (ATM) activation and caused growth inhibition in MCF-7 cells. It also improved the radiosensitivity of MCF-7 cells (Table 1) [85].

### AZD8055

AZD8055 is a potent ATP-competitive inhibitor of mTORC1 and mTOR2 with an  $IC_{50}$  value of 0.8 nM (Fig. 2). Chang et al. evaluated the effect of AZD8055 on radiosensitivity of nasopharyngeal carcinoma (NPC) cells. They suggested that the combination of AZD8055 with radiation markedly enhanced the inhibitions of both cell proliferation and clonogenicity. In contrast to the application of ionizing radiation alone, AZD8055 combined with 4 Gy radiation resulted in

2.63-fold and 3.62-fold reductions in the survival fractions (SF) of the CNE1 and CNE2 cells respectively. Combining IR with AZD8055 delayed tumor growth and induced considerably far more apoptosis in comparison with IR and AZD8055 alone. The results displayed that 5 mg/kg/d AZD8055 induced good radiosensitivity for NPC (Table 1) [86].

### SF1126

SF1126 is a peptidic pro-drug inhibitor of both PI3 kinase and mTOR. Shu et al. evaluated the effect of SF1126 on Glioma cells. All glioma cells (SF767, LNpuro, and LNvIII) displayed considerable improvement of radiosensitivity after treatment with SF1126. Moreover, SF1126 demonstrated direct effects on glioma tumor cells. SF1126 significantly inhibited tumor growth (Table 1) [87].

### 3-Methyladenine

3-Methyladenine (3-MA) is a PI3K autophagy inhibitor (Fig. 2). Chen et al. studied the effects of 3-MA on radiosensitivity of EC9706 cells. 3-MA impressively inhibited the autophagy induced by radiation therapy. Autophagy inhibition enhanced cell apoptosis and reduced tumor proliferation and resulted in a significant increase in the radio-susceptibility of the tumor cells (Table 1) [88].

### LY294002

LY294002 (2-(2-morpholinoethyl)-8-phenylchromone) is a cell-permeable inhibitor of PI3Ks (Fig. 2). It is reported that LY294002 as an inhibitor of PI3K sensitizes the PTEN mutant glioma cell line U251 MG to radiation [89]. Kao et al. evaluated the effect of LY294002 on the radiosensitivity of U251 cells. Inhibition of PI3K signaling pathway by LY294002 treatment resulted in suppression of DNA repair, which led to tumor cell death. Hence, it made the cells more sensitive to ionizing radiation (Fig. 2; Table 1) [90]. It is noteworthy that the toxicity of LY294002 makes it unfavorable for clinical trials [91]. Another study tested the effect of LY294002 in combination with irradiation on eight GBM cell lines. Among these cell lines only U87MG, MO59J, and LN18 display increased phosphorylation of AKT after irradiation (single dose, 6 Gy). The treatment of LY294002 before irradiation completely abolishes phosphorylation of AKT in the U87-MG cell line [92]. A combination of LY294002, gemcitabine hydrochloride, and ionizing radiation effectively inhibited vasculogenic mimicry formation and matrix metalloproteinase-2 mRNA expression of Panc-1 cells line. This combination was also effective against tumor growth in orthotopic Panc-1 xenografts by suppressing PI3K/MMPs/Ln-5 $\gamma$ 2 signaling pathway [93].

### PI-103

PI-103 is a potent ATP-competitive inhibitor of PI3K/AKT and mTOR (Fig. 2) [94]. Prevo et al. investigated the effect of PI-103 on the radio-sensitivity of HCT116 and DLD1 cancer cells. Findings revealed that the inhibition of PI3K causes a reduction in AKT phosphorylation. AKT activation played a major role in cell survival reduction. PI-103 combined with radiation increased delay in the G2/M phase. PI-103 was found to improve radiation therapy efficacy and radiation-induced apoptosis (Table 1). PI-103 prolongs DNA DSB repair by raising the levels of  $\gamma$ -H2AX and reduces cell proliferation [95]. A similar study on colon carcinoma cell lines (SW480 and SW48) revealed that treating these cell lines with a combination of PI-103 and NVP-AUY922 (Heat Shock Protein 90 (HSP-90) inhibitor) three hours ahead of radiotherapy (the treatment was continued for 24 h) effectively increases the radiosensitivity [96].

### Omipalisib (GSK2126458)

Omipalisib is a potent and selective inhibitor of PI3K/mTOR (Fig. 2). Liu et al. evaluated the effect of both omipalisib and gedatolisib on the radiosensitivity of NPC cells. Omipalisib and gedatolisib combined with ionizing radiation effectively inhibited the proliferation of NPC cells. Both inhibitors increased the radiosensitivity of cells by DNA damage enhancement, delaying the G2/M phases, and enhanced cell apoptosis. The combination of omipalisib or gedatolisib with radiation considerably induced inhibition of tumor growth (Table 1) [97].

### Palomid-529

Palomid-529 (8-(1-Hydroxy-ethyl)-2-methoxy-3-(4-methoxy-benzyloxy)-benzo[*c*]chromen-6-one) (Fig. 2), inhibits both TORC1 and TORC2. It also blocks phosphorylation of AKT at S473 and reduces tumor growth, angiogenesis, and vascular permeability in the xenograft mouse model of glioblastoma (GB) [98]. A study indicated that palomid-529 possesses anti-tumor activity against a large variety of cell lines (NCI-60 cell lines panel). This compound increased the radiosensitivity of NCI-60 cell lines panel and reduced the level of phosphorylated AKT (pAKT), matrix metalloproteinase (MMP)-2, and VEGF. The treatment of the PC-3 xenograft model with a combination of palomid-529 (20 mg/Kg) and radiation (6 Gy) led to 77.4% tumor shrinkage while using palomid-529 (20 mg/Kg) or radiation (6 Gy) shrank the tumor size by 42.9 and 53 %, respectively (Table 1) [99].

### HS-173

Ethyl 6-(5-(phenylsulfonamido)pyridin-3-yl)imidazo[1,2-*a*]pyridine-3-carboxylate (HS-173) (Fig. 2), is a new PI3K $\alpha$

inhibitor. HS-173 in combination with radiation inhibits radiation-induced DNA breaks repair, G2/M cell cycle arrest, and apoptosis in Miapaca-2 and PANC-1 pancreatic cell lines (Table 1) [100].

### Conclusion

Radiotherapy reduces both tumor size and cancer recurrence. However, due to the radioresistance of several cancer cell lines, effective cancer treatment utilizes suitable adjuvants. Adjuvants utilized in radiation therapy should increase the radiosensitivity of tumor cells. Cancer cells might be resistant to apoptosis especially through PI3K/AKT/mTOR signaling pathway. Therefore, mTOR pathway inhibition could impressively weaken radioresistance and make cancer cells supersensitive to apoptosis. Noteworthy, dual inhibitors of PI3K/AKT/mTOR signaling pathway seem to be superior to single component inhibitors in radiosensitizing different cell lines. Hence, the combination of such inhibitors with radiation can be considered as more effective adjuvants to increase the sensitivity of cancer cells to radiation.

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### References

1. Thorpe LM, Yuzugullu H, Zhao JJ (2015) PI3K in cancer: divergent roles of isoforms, modes of activation and therapeutic targeting. *Nat Rev Cancer* 15(1):7–24
2. Guertin DA, Sabatini DM (2007) Defining the role of mTOR in cancer. *Cancer Cell* 12(1):9–22
3. Menon S, Manning BD (2008) Common corruption of the mTOR signaling network in human tumors. *Oncogene* 27(2):S43–S51
4. Zaytseva YY et al (2012) mTOR inhibitors in cancer therapy. *Cancer Lett* 319(1):1–7
5. Pedroza DA et al (2020) Progesterone receptor membrane component 1 promotes the growth of breast cancers by altering the phosphoproteome and augmenting EGFR/PI3K/AKT signalling. *Br J Cancer* 123:1326–1335

6. Markova B et al (2010) Novel pathway in Bcr-Abl signal transduction involves Akt-independent, PLC- $\gamma$ 1-driven activation of mTOR/p70S6-kinase pathway. *Oncogene* 29(5):739–751
7. Cornu M, Albert V, Hall MN (2013) mTOR in aging, metabolism, and cancer. *Curr Opin Genet Dev* 23(1):53–62
8. Zhou H, Luo Y, Huang S (2010) Updates of mTOR inhibitors. *Anti-Cancer Agents Med Chem* 10(7):571–581
9. Francipane MG, Lagasse E (2014) mTOR pathway in colorectal cancer: an update. *Oncotarget* 5(1):49
10. Bellizzi AM et al (2010) The mTOR pathway is frequently activated in pancreatic ductal adenocarcinoma and chronic pancreatitis. *Appl Immunohistochem Mol Morphol* 18(5):442–447
11. Li X et al (2016) PI3K/Akt/mTOR signaling pathway and targeted therapy for glioblastoma. *Oncotarget* 7(22):33440
12. Hütt-Cabezas M et al (2013) Activation of mTORC1/mTORC2 signaling in pediatric low-grade glioma and pilocytic astrocytoma reveals mTOR as a therapeutic target. *Neurooncology* 15(12):1604–1614
13. Bilanges B, Posor Y, Vanhaesebroeck B (2019) PI3K isoforms in cell signalling and vesicle trafficking. *Nat Rev Mol Cell Biol* 20(9):515–534
14. Lee Y-R, Chen M, Pandolfi PP (2018) The functions and regulation of the PTEN tumour suppressor: new modes and prospects. *Nat Rev Mol Cell Biol* 19(9):547–562
15. Hoxhaj G, Manning BD (2020) The PI3K–AKT network at the interface of oncogenic signalling and cancer metabolism. *Nat Rev Cancer* 20(2):74–88
16. Valvezan AJ, Manning BD (2019) Molecular logic of mTORC1 signalling as a metabolic rheostat. *Nat Metab* 1(3):321–333
17. Chen X et al (2018) Cryo-EM structure of human mTOR complex 2. *Cell Res* 28(5):518–528
18. Dibble CC, Manning BD (2013) Signal integration by mTORC1 coordinates nutrient input with biosynthetic output. *Nat Cell Biol* 15(6):555–564
19. Bracho-Valdés I et al (2011) mTORC1-and mTORC2-interacting proteins keep their multifunctional partners focused. *IUBMB Life* 63(10):896–914
20. Bond P (2016) Regulation of mTORC1 by growth factors, energy status, amino acids and mechanical stimuli at a glance. *J Int Soc Sports Nutr* 13(1):8
21. Showkat M, Beigh MA, Andrabi KI (2014) mTOR signaling in protein translation regulation: implications in cancer genesis and therapeutic interventions. *Mol Biol Int* 2014:1–14
22. Oh WJ, Jacinto E (2011) mTOR complex 2 signaling and functions. *Cell Cycle* 10(14):2305–2316
23. Kaizuka T et al (2010) Tti1 and Tel2 are critical factors in mammalian target of rapamycin complex assembly. *J Biol Chem* 285(26):20109–20116
24. Chang L et al (2015) Targeting PI3K/Akt/mTOR signaling pathway in the treatment of prostate cancer radioresistance. *Crit Rev Oncol Hematol* 96(3):507–517
25. Edwards E et al (2002) Phosphatidylinositol 3-kinase/Akt signaling in the response of vascular endothelium to ionizing radiation. *Cancer Res* 62(16):4671–4677
26. Schmidt-Ullrich RK et al (2003) ERBB receptor tyrosine kinases and cellular radiation responses. *Oncogene* 22(37):5855–5865
27. Zoncu R, Efeyan A, Sabatini DM (2011) mTOR: from growth signal integration to cancer, diabetes and ageing. *Nat Rev Mol Cell Biol* 12(1):21–35
28. Wang L et al (2016) Autophagy inhibition sensitizes WYE-354-induced anti-colon cancer activity in vitro and in vivo. *Tumor Biol* 37(9):11743–11752
29. Pan XD et al (2017) Concurrent inhibition of mTORC1 and mTORC2 by WYE-687 inhibits renal cell carcinoma cell growth in vitro and in vivo. *PLoS ONE*. <https://doi.org/10.1371/journal.pone.0172555>
30. Xing X et al (2014) PP242 suppresses cell proliferation, metastasis, and angiogenesis of gastric cancer through inhibition of the PI3K/AKT/mTOR pathway. *Anti-Cancer Drugs* 25(10):1129
31. Huang H et al (2017) Two mTOR inhibitors, rapamycin and Torin 1, differentially regulate iron-induced generation of mitochondrial ROS. *Biomaterials* 30(6):975–980
32. Yongxi T et al (2015) Autophagy inhibition sensitizes KU-0063794-mediated anti-HepG2 hepatocellular carcinoma cell activity in vitro and in vivo. *Biochem Biophys Res Commun* 465(3):494–500
33. Yu K et al (2009) Biochemical, cellular, and in vivo activity of novel ATP-competitive and selective inhibitors of the mammalian target of rapamycin. *Cancer Res* 69(15):6232–6240
34. Garcia-Echeverria C (2011) Blocking the mTOR pathway: a drug discovery perspective. Portland Press Ltd, London
35. Zhou H-Y, Huang S-L (2012) Current development of the second generation of mTOR inhibitors as anticancer agents. *Chin J Cancer* 31(1):8
36. Mayer IA, Arteaga CL (2016) The PI3K/AKT pathway as a target for cancer treatment. *Ann Rev Med* 67:11–28
37. Rizzieri DA et al (2008) A phase 2 clinical trial of deforolimus (AP23573, MK-8669), a novel mammalian target of rapamycin inhibitor, in patients with relapsed or refractory hematologic malignancies. *Clin Cancer Res* 14(9):2756–2762
38. Sinclair C et al (2017) mTOR regulates metabolic adaptation of APCs in the lung and controls the outcome of allergic inflammation. *Science* 357(6355):1014–1021
39. Eynott PR et al (2003) Effects of cyclosporin A and a rapamycin derivative (SAR943) on chronic allergic inflammation in sensitized rats. *Immunology* 109(3):461–467
40. Chibana H et al (2017) Interleukin-1 $\beta$  is associated with coronary endothelial dysfunction in patients with mTOR-inhibitor-eluting stent implantation. *Heart Vessels* 32(7):823–832
41. Rini BI (2008) Temsirolimus, an inhibitor of mammalian target of rapamycin. *Clin Cancer Res* 14(5):1286–1290
42. Liu Q et al (2009) mTOR mediated anti-cancer drug discovery. *Drug Discov Today Ther Strateg* 6(2):47–55
43. Alvarado Y et al (2011) Clinical activity of mammalian target of rapamycin inhibitors in solid tumors. *Target Oncol* 6(2):69–94
44. Tian T, Li X, Zhang J (2019) mTOR signaling in cancer and mTOR inhibitors in solid tumor targeting therapy. *Int J Mol Sci* 20(3):755
45. Tam SY, Wu VWC, Law HKW (2017) Influence of autophagy on the efficacy of radiotherapy. *Radiat Oncol* 12(1):57
46. Yoo YJ et al (2017) An overview of rapamycin: from discovery to future perspectives. *J Ind Microbiol Biotechnol* 44(4–5):537–553
47. Weppeler SA et al (2007) Response of U87 glioma xenografts treated with concurrent rapamycin and fractionated radiotherapy: possible role for thrombosis. *Radiation Oncol* 82(1):96–104
48. Dumont RA et al (2013) Targeted radiotherapy of prostate cancer with a gastrin-releasing peptide receptor antagonist is effective as monotherapy and in combination with rapamycin. *J Nucl Med* 54(5):762–769
49. Eshleman JS et al (2002) Inhibition of the mammalian target of rapamycin sensitizes U87 xenografts to fractionated radiation therapy. *Cancer Res* 62(24):7291–7297
50. Anandharaj A, Cinghu S, Park W-Y (2011) Rapamycin-mediated mTOR inhibition attenuates survivin and sensitizes glioblastoma cells to radiation therapy. *Acta Biochim Biophys Sin* 43(4):292–300
51. Buijssen J et al (2015) A phase I–II study on the combination of rapamycin and short course radiotherapy in rectal cancer. *Radiation Oncol* 116(2):214–220
52. Dancey JE, Curiel R, Purvis J (2009) Evaluating temsirolimus activity in multiple tumors: a review of clinical trials. Elsevier, Amsterdam

53. Ekshyyan O et al (2009) Comparison of radiosensitizing effects of the mammalian target of rapamycin inhibitor CCI-779 to cisplatin in experimental models of head and neck squamous cell carcinoma. *Mol Cancer Ther* 8(8):2255–2265
54. Du Y, Peyser ND, Grandis JR (2014) Integration of molecular targeted therapy with radiation in head and neck cancer. *Pharmacol Ther* 142(1):88–98
55. Ushijima H et al (2015) Radio-sensitization effect of an mTOR inhibitor, temsirolimus, on lung adenocarcinoma A549 cells under normoxic and hypoxic conditions. *J Radiat Res* 56(4):663–668
56. Cao C et al (2006) Inhibition of mammalian target of rapamycin or apoptotic pathway induces autophagy and radiosensitizes PTEN null prostate cancer cells. *Cancer Res* 66(20):10040–10047
57. Yu C-C et al (2014) RAD001 enhances the radiosensitivity of SCC4 oral cancer cells by inducing cell cycle arrest at the G2/M checkpoint. *Anticancer Res* 34(6):2927–2935
58. Kim KW et al (2008) Autophagy upregulation by inhibitors of caspase-3 and mTOR enhances radiotherapy in a mouse model of lung cancer. *Autophagy* 4(5):659–668
59. Gelsomino F et al (2017) A Dose-finding and biomarker evaluation phase Ib study of everolimus in association with 5-fluorouracil and pelvic radiotherapy as neoadjuvant treatment of locally advanced rectal cancer (E-LARC Study). *Clin Colorectal Cancer* 16(4):410–415.e1
60. Azria D et al (2013) Concurrent treatment with everolimus (RAD001) and hormonoradiotherapy in high-risk locally advanced prostate cancer: results of a phase I trial. *J Clin Oncol* 31(6\_suppl):150–150
61. Narayan V et al (2017) Phase I trial of everolimus and radiation therapy for salvage treatment of biochemical recurrence in prostate cancer patients following prostatectomy. *Int J Radiat Oncol Biol Phys* 97(2):355–361
62. de Melo AC et al (2016) A phase I study of mTOR inhibitor everolimus in association with cisplatin and radiotherapy for the treatment of locally advanced cervix cancer: PHOENIX I. *Cancer Chemother Pharmacol* 78(1):101–109
63. Sarkaria JN et al (2011) North central cancer treatment group phase I trial N057K of everolimus (RAD001) and temozolomide in combination with radiation therapy in patients with newly diagnosed glioblastoma multiforme. *Int J Radiat Oncol Biol Phys* 81(2):468–475
64. Ma DJ et al (2015) A phase II trial of everolimus, temozolomide, and radiotherapy in patients with newly diagnosed glioblastoma: NCCTG N057K. *Neuro Oncol* 17(9):1261–1269
65. Chinnaiyan P et al (2018) A randomized phase II study of everolimus in combination with chemoradiation in newly diagnosed glioblastoma: results of NRG oncology RTOG 0913. *Neuro Oncol* 20(5):666–673
66. Deutsch E et al (2015) Phase I trial of everolimus in combination with thoracic radiotherapy in non-small-cell lung cancer. *Ann Oncol* 26(6):1223–1229
67. Prasad G et al (2011) Inhibition of PI3K/mTOR pathways in glioblastoma and implications for combination therapy with temozolomide. *Neuro Oncol* 13(4):384–392
68. Wen PY et al (2015) Phase I dose-escalation study of the PI3K/mTOR inhibitor voxalisib (SAR245409, XL765) plus temozolomide with or without radiotherapy in patients with high-grade glioma. *Neuro Oncol* 17(9):1275–1283
69. Gil del Alcazar, CR et al (2014) Inhibition of DNA double-strand break repair by the dual PI3K/mTOR inhibitor NVP-BE2235 as a strategy for radiosensitization of glioblastoma. *Clin Cancer Res* 20(5):1235–1248
70. Cerniglia GJ et al (2012) Inhibition of autophagy as a strategy to augment radiosensitization by the dual phosphatidylinositol 3-kinase/mammalian target of rapamycin inhibitor NVP-BE2235. *Mol Pharmacol* 82(6):1230–1240
71. Gao M et al (2012) Dual PI3K/mTOR blockade by NVP-BE2235 sensitizes head and neck squamous cell carcinoma to radiotherapy. *Cancer Res*. <https://doi.org/10.1158/1538-7445>
72. Chen YH et al (2015) Dual phosphoinositide 3-kinase/mammalian target of rapamycin inhibitor is an effective radiosensitizer for colorectal cancer. *Cancer Lett* 357(2):582–590
73. Chen Y-H et al (2019) Maintenance BEZ235 treatment prolongs the therapeutic effect of the combination of BEZ235 and radiotherapy for colorectal cancer. *Cancers* 11(8):1204
74. Chang L et al (2014) PI3K/Akt/mTOR pathway inhibitors enhance radiosensitivity in radioresistant prostate cancer cells through inducing apoptosis, reducing autophagy, suppressing NHEJ and HR repair pathways. *Cell Death Dis* 5(10):e1437–e1437
75. Potiron VA et al (2013) Radiosensitization of prostate cancer cells by the dual PI3K/mTOR inhibitor BEZ235 under normoxic and hypoxic conditions. *Radiother Oncol* 106(1):138–146
76. Zhu W, Fu W, Hu L (2013) NVP-BE2235, dual phosphatidylinositol 3-kinase/mammalian target of rapamycin inhibitor, prominently enhances radiosensitivity of prostate cancer cell line PC-3. *Cancer Biother Radiopharm* 28(9):665–673
77. Kuger S et al (2014) Novel PI3K and mTOR inhibitor NVP-BE2235 radiosensitizes breast cancer cell lines under normoxic and hypoxic conditions. *Breast Cancer* 8:39–49
78. Wang WJ et al (2013) NVP-BE2235, a novel dual PI3K/mTOR inhibitor, enhances the radiosensitivity of human glioma stem cells in vitro. *Acta Pharmacol Sin* 34(5):681–690
79. Kuger S et al (2013) Radiosensitization of glioblastoma cell lines by the dual PI3K and mTOR inhibitor NVP-BE2235 depends on drug-irradiation schedule. *Transl Oncol* 6(2):169–1N16
80. Leiker AJ et al (2015) Radiation enhancement of head and neck squamous cell carcinoma by the dual PI3K/mTOR inhibitor PF-05212384. *Clin Cancer Res* 21(12):2792–2801
81. Wilson GD et al (2021) Dacomitinib and gedatolisib in combination with fractionated radiation in head and neck cancer. *Clin Transl Radiat Oncol* 26:15–23
82. Kahn J et al (2014) The mTORC1/mTORC2 inhibitor AZD2014 enhances the radiosensitivity of glioblastoma stem-like cells. *Neurooncology* 16(1):29–37
83. Hayman TJ et al (2014) The ATP-competitive mTOR inhibitor INK128 enhances in vitro and in vivo radiosensitivity of pancreatic carcinoma cells. *Clin Cancer Res* 20(1):110–119
84. Connolly E et al (2011) TORC1/2 inhibition with concurrent radiation controls inflammatory breast cancer in a preclinical animal model through selective blockade of translation. *Int J Radiat Oncol Biol Phys* 81(2):S751
85. Luo J et al (2018) Torin2 enhances the radiosensitivity of MCF-7 breast cancer cells by downregulating the mTOR signaling pathway and ATM phosphorylation. *Mol Med Rep* 17(1):366–373
86. Chang L et al (2019) A low dose of AZD8055 enhances radiosensitivity of nasopharyngeal carcinoma cells by activating autophagy and apoptosis. *Am J Cancer Res* 9(9):1922
87. Shu H et al (2007) SF1126, a novel integrin-targeting phosphatidylinositol-3 kinase inhibitor, has radiosensitizing and anti-tumor effects in glioma model systems. *Int J Radiat Oncol Biol Phys* 69(3):S98
88. Chen Y et al (2015) Combining radiation with autophagy inhibition enhances suppression of tumor growth and angiogenesis in esophageal cancer. *Mol Med Rep* 12(2):1645–1652
89. Nakamura JL et al (2005) PKB/Akt mediates radiosensitization by the signaling inhibitor LY294002 in human malignant gliomas. *J Neurooncol* 71(3):215–222

90. Kao GD et al (2007) Inhibition of phosphatidylinositol-3-OH kinase/Akt signaling impairs DNA repair in glioblastoma cells following ionizing radiation. *J Biol Chem* 282(29):21206–21212
91. Gupta AK et al (2003) Radiation sensitization of human cancer cells in vivo by inhibiting the activity of PI3K using LY294002. *Int J Radiat Oncol Biol Phys* 56(3):846–853
92. Li HF, Kim JS, Waldman T (2009) Radiation-induced Akt activation modulates radioresistance in human glioblastoma cells. *Radiat Oncol* 4:43
93. Bai R et al (2016) The effect of PI3K inhibitor LY294002 and gemcitabine hydrochloride combined with ionizing radiation on the formation of vasculogenic mimicry of Panc-1 cells in vitro and in vivo. *Neoplasma* 63(1):80–92
94. Raynaud FI et al (2007) Pharmacologic characterization of a potent inhibitor of class I phosphatidylinositide 3-kinases. *Cancer Res* 67(12):5840–5850
95. Prevo R et al (2008) Class I PI3 kinase inhibition by the pyridinylfuranopyrimidine inhibitor PI-103 enhances tumor radiosensitivity. *Cancer Res* 68(14):5915–5923
96. Djuzenova CS et al (2016) Dual PI3K- and mTOR-inhibitor PI-103 can either enhance or reduce the radiosensitizing effect of the Hsp90 inhibitor NVP-AUY922 in tumor cells: the role of drug-irradiation schedule. *Oncotarget* 7(25):38191–38209
97. Liu T et al (2015) Dual PI3K/mTOR inhibitors, GSK2126458 and PKI-587, suppress tumor progression and increase radiosensitivity in nasopharyngeal carcinoma. *Mol Cancer Ther* 14(2):429–439
98. Xue Q et al (2008) Palomid 529, a novel small-molecule drug, is a TORC1/TORC2 inhibitor that reduces tumor growth, tumor angiogenesis, and vascular permeability. *Cancer Res* 68(22):9551–9557
99. Diaz R et al (2009) The novel Akt inhibitor palomid 529 (P529) enhances the effect of radiotherapy in prostate cancer. *Br J Cancer* 100(6):932–940
100. Park JH et al (2017) Radiosensitization of the PI3K inhibitor HS-173 through reduction of DNA damage repair in pancreatic cancer. *Oncotarget* 8(68):112893–112906

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