

Review Article

Targeting autophagy in chemotherapy-resistant of hepatocellular carcinoma

Jiyao Sheng², Hanjiao Qin³, Kun Zhang¹, Bingjin Li¹, Xuewen Zhang²

¹Jilin Provincial Key Laboratory on Molecular and Chemical Genetic, Departments of ²Hepatobiliary and Pancreatic Surgery, ³Radiotherapy, The Second Hospital of Jilin University, Changchun 130041, Jilin, China

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Abstract: Hepatocellular carcinoma (HCC) is a malignant tumor with poor prognosis. Surgical resection is recommended for very early-stage and early-stage HCC, but HCC is still prone to recurrence and metastasis after surgery. Furthermore, treatment options for intermediate- and advanced-stage HCC are relatively limited. Systemic therapy is the preferred method to kill residual cancer cells after surgery and prolong survival time of inoperable patients, but most cases are insensitive to chemotherapeutic agents, restricting widespread clinical application of systemic therapy. Many studies have found that various chemotherapeutic drugs for HCC treatment can increase autophagic flux of HCC cells, and it may be related with enhancing drug resistance and promoting cell survival. However, enhancement of autophagic flux may also induce tumor cell death in some cases, leading to marked inconsistency across studies. Here we reviewed the mechanisms underlying the increase in autophagic flux in HCC cells induced by chemotherapeutic drugs and examined the contributions of autophagy and related pathways to chemotherapy drug resistance. Our aim was to identify potential autophagy-related targets for improving the sensitivity of HCC to chemotherapeutic drugs.

Keywords: Hepatocellular carcinom, chemotherapy-resistant, autophagy, cancer

Introduction

Hepatocellular carcinoma (HCC) is the most common primary liver cancer. According to the World Health Organization (WHO), HCC is the fifth most common tumor worldwide and the second most common cause of cancer-related death [1]. At present, the main therapeutic options for HCC are surgical resection, liver transplantation, ablation, arterial directed therapy, radiotherapy, and systemic therapy. According to the Barcelona Clinic Liver Cancer (BCLC) classification and treatment strategy, surgical resection is the most effective choice for improving the prognosis of very early-stage (0) and early-stage (A) HCC patients, with a reported 5-year survival after resection of about 50%-70% [2-4]. However, HCC demonstrates strong invasive and metastatic capacities, allowing HCC cells to disseminate from the primary tumor before or during surgical resection [5]. In addition, most HCC cases result from progression of cirrhosis, and cells of

the cirrhotic liver may undergo tumorigenesis and form de novo tumors after surgical resection [6], resulting in high postoperative HCC recurrence (>70% at 5 years) [7]. Therefore, effective systemic therapy during the perioperative period can decrease the recurrence rate and improves prognosis. In patients with intermediate- or advanced-stage HCC, however, intrahepatic or distant metastasis may have already occurred. In such cases, prognosis is poor and treatment options are limited, therefore, more effective systemic treatments are needed to slow the progression of HCC.

Chemotherapy, including traditional chemotherapeutic agents and small-molecule inhibitors, is the most widely used systemic therapy for cancer and is a pivotal component of comprehensive postoperative HCC treatment for the prevention of tumor recurrence and treatment of inoperable patients [8]. However, HCC cells have low sensitivity to traditional chemotherapeutic agents which leads a poor efficacy.

Chemosensitization of HCC by targeting autophagy

Although a multicenter randomized controlled trial (RCT) in Asia indicated that an oxaliplatin-based FOLFOX4 chemotherapy regimen is superior to doxorubicin with respect to the overall response rate, disease control rate, progression-free survival, and overall survival [9]. However, many hepatobiliary cancer guidelines do not recommend systemic chemotherapy as the first-line treatment [10-13] due to the relatively low sensitivity of HCC cells. Trans-arterial chemoembolization (TACE), the intra-arterial infusion of a higher dose of traditional chemotherapy agent embedded in lipiodol as a vehicle for increasing tumour exposure to the drug, followed embolization in arterial flow of hepatic arterial supply of HCC. It has been the most effective therapy to prolong survival time of patients with advanced HCC who lost the opportunity for operation and widely used in clinical treatment. Nearly half of all HCC patients undergo this procedure at some point during treatment [14]. TACE can prolong the action time of high-concentration chemotherapeutic drugs while reducing toxicity to non-target tissues. In an RCT including 80 patients, the overall survival of the TACE group was significantly improved compared with that of the control group administered only symptomatic treatment (1 year, 57% vs. 32%; 2 years, 31% vs. 11%; 3 years, 26% vs. 3%; $P=0.002$) [15]. Other clinical studies have also concluded that TACE can prolong survival of patients with advanced-stage HCC [16-18]. Therefore, TACE is recommended as an effective therapy for unresectable HCC by various international guidelines [10-13]. However, there are outstanding questions worthy of further study, such as how well various chemotherapy regimens work in TACE and whether it is possible to prolong survival by increasing the sensitivity of HCC cells to chemotherapeutic drugs.

In addition to TACE, small-molecule inhibitors such as sorafenib have shown promise for the clinical treatment of HCC. Sorafenib is a multikinase inhibitor which can inhibit multiple tyrosine kinases involved in tumor proliferation, angiogenesis, and apoptosis, such as vascular endothelial growth factor receptor (VEGFR), platelet-derived growth factor receptor (PDGFR), and Raf family kinases [19]. Sorafenib is the only Food and Drug Administration (FDA)-approved HCC systemic therapy shown to significantly prolong overall survival of patients

with advanced-stage HCC [20, 21]. However, only approximately 30% of patients are responsive to sorafenib and ultimately benefit from it, moreover, most patients eventually develop resistance to the drug and have a poor prognosis [21, 22]. Therefore, improving the sensitivity of HCC cells to both traditional chemotherapeutic agents and small-molecule inhibitors is critical for prolonging survival.

Macroautophagy (hereafter referred to as autophagy) is an ancient and highly conserved catabolic process during evolution, when cells are in starvation or stress state, intracellular proteins and organelles can be engulfed by double-membrane vesicles known as autophagosomes, and then delivery to lysosomes for degradation to biosynthetic precursors and energy sources. Autophagy occurs at a basal level in cells and can be induced by diverse signals and cellular stressors including treatment of chemotherapeutic agents. Intensive study of autophagy has revealed that this process can act as both a tumor suppressor and a protector of cancer cells depending on factors such as the degree of induction (autophagic flux) and stage of tumor progression. In some cases, autophagy can remove genomic instability, damaged organelles and mutated cells in the early stage of tumor formation, thereby inhibiting tumorigenesis [23, 24]. However, cells in established tumors can resist the damage of chemotherapeutic drugs through autophagy to indeed, autophagy is an important defense mechanism of tumor cells against chemotherapeutic drugs. The cellular components damaged by chemotherapeutic drugs (including misfolded proteins, damaged DNA and damaged organelles) can be degraded to substances that support metabolism and promote further tumor cell growth and survival through autophagy. The resistance mechanism associated autophagy can be divided into two cases, some kinds of tumor cell types maintain high basal autophagic flux, resulting in intrinsic resistance to chemotherapeutic drugs while others gradually acquire drug resistance by increasing autophagic flux in response to the treatment of chemotherapeutic drugs, thus, autophagy underlies a type of acquired drug resistance. However, enhancement of autophagic flux may also induce tumor cell death in some cases, leading to marked inconsistency across studies. In brief, a better understand-

Chemosensitization of HCC by targeting autophagy

Table 1. Role of key molecules of the autophagic pathway and related pathways in resistance of hepatocellular carcinoma to chemotherapy

Chemotherapy drug	Autophagy related targets	Combination therapy	Model	Role of autophagy	Renfereces
Oxaliplatin	Lysosome	Oxaliplatin+CQ	HepG2, Huh7, SMMC-7721	Cell survival	[26, 46]
	ATG 7	Oxaliplatin+ATG7 siRNA	HepG2	Cell survival	[26]
Cisplatin	Lysosome	Cisplatin+CQ	HepG2	Cell survival	[29]
	PI3K III	Cisplatin+3-MA	HepG2	Cell survival	[29]
	ATG4D	Cisplatin+miR-101	HepG2	Cell survival	[28]
Sorafenib	ATG 5	Sorafenib+ATG5 siRNA	MHCC97-L, PLC/PRF/5	Cell survival	[30]
	Lysosome	Sorafenib+CQ	MHCC97-L, PLC/PRF/5, Huh7, Bel-7402, Hep3B	Cell survival	[30-32]
	Lysosome	Sorafenib+CQ/Bafilomycin A1	PLC/PRF/5, SK-Hep1	Cell death	[33]
	PI3K III	Sorafenib+3MA	MHCC97-L, PLC/PRF/5, Bel-7402, Hep3B	Cell survival	[30, 32]
	ATG 7	Sorafenib+ATG7 siRNA	Huh7, PLC/PRF/5	Cell survival	[31]
	HIF-1 α	Sorafenib+melatonin	Hep3B	Cell survival	[90]
Doxorubicin	Autophagosome and lysosome	Doxorubicin+20(S)-ginsenoside Rg3	SK-Hep1, HepG2, Mouse model	Cell survival	[25]
	PI3K III	Doxorubicin+3-MA	Hep3B	Cell survival	[47]
	mTOR	Doxorubicin+rapamycin	Hep3B	Cell survival	[47]
	mTOR	Doxorubicin+rapamycin+miR-26	HepG2	Cell survival	[69]
	Akt	Doxorubicin+MK-2206	SNU449, SNU475	Cell survival	[78]

Note: ATG, Autophagy-related protein; PI3K III, Phosphatidylinositol 3-kinase Class III; HIF-1 α , Hypoxia-inducible factor-1-alpha; mTOR, mammalian target of rapamycin; CQ, chloroquine; 3-MA, 3-Methyladenine.

ing of the role of autophagy in HCC will help in the understanding of the mechanisms of chemotherapy resistance.

Autophagy and chemoresistance of HCC

Chemotherapeutic drugs have been shown to induce autophagy in HCC cells both in vitro and in vivo, as shown in **Table 1**. Kim et al found that doxorubicin-induced dose- and time-dependent autophagy in HCC cells. The steroidal saponin 20(S)-Ginsenoside Rg3 inhibited autophagic flux by suppressing late-stage autophagosome maturation and lysosomal degradation. Moreover, 20(S)-Ginsenoside Rg3 inhibited doxorubicin-induced autophagy and sensitized HCC cells to doxorubicin-induced cell death in vitro and in vivo. Of critical importance for cancer therapy is that 20(S)-Ginsenoside Rg3 did not enhance the sensitivity of normal human liver cells to doxorubicin, suggesting that HCC is specifically targeted for destruction through inhibition of autophagy [25]. Like doxorubicin, oxaliplatin-induced autophagy in HCC cell lines, and their sensitivity to oxaliplatin was significantly enhanced following autophagy inhibition by treatment with chloroquine (CQ) or a small interfering RNA (siRNA) against the autophagy-related protein Atg7 [26]. In Huh7 and HepG2 cells, cisplatin increased the ratio of the autophagosome-associated protein LC3-II (a phosphatidylethanolamine-conjugated form of microtubule associated protein 1A/B light chain 3) to its non-lipidated cytosolic precursor LC3-I (LC3-II/LC3-I), a widely used marker for autophagic flux, and promoted the degradation of sequestosome-1 (SQSTM1 or ubiquitin-binding protein p62), an autophagosome protein that targets specific proteins for selective autophagic degradation. Thus, like doxorubicin and oxaliplatin, cisplatin appears to increase autophagic flux in HCC cells [27]. Other studies, including those by the author's team, have found that different autophagy inhibitors can increase the sensitivity of HCC cells to cisplatin [28, 29]. Furthermore, sorafenib can induce autophagy in various hepatocellular carcinoma cell lines (Huh7, HLF, PLC/PRF/5, Hep3B, Sk-Hep1, MHCC97-L, and HepG2) [30-33], possibly by promoting lipidation of LC3-I and the expression of key autophagy molecules such as Beclin-1, ATG5, ATG7, and LC3 [34]. However, the role of autophagy in drug resistance of HCCs is still controversial.

Although several studies have shown that autophagy contributes to the resistance of HCC cells to sorafenib, because the combination of autophagy inhibitors or silencing of key autophagy genes can enhance the cytotoxic effect of sorafenib on HCC cells [30-32]. But a cell death-promoting effect of sorafenib-induced autophagy has also been reported by several studies [33], this may be related that the sorafenib can induce autophagic death of HCC.

The hypoxic state of tumor cells also contributes to the induction of autophagy, and HCC is one of the most hypoxic of all tumors, with a median oxygen level as low as 0.8% [35]. In the course of chemotherapy, sorafenib can inhibit tumor angiogenesis, thereby reducing tumor cell blood supply and exacerbating HCC hypoxia. The nutrient arteries of HCC are also embolized during TACE, aggravating tumor hypoxia [36, 37]. The hypoxic state of HCC is also one reason for constitutive upregulation of autophagic flux [38].

In addition, the key molecules of autophagy may affect HCC sensitivity to chemotherapeutic drugs by interacting with apoptosis-related proteins. Zhou et al found that autophagy contributes to the chemoresistance of HCC cells by downregulating proapoptotic proteins such as Bad and Bim, resulting in suppression of mitomycin-induced apoptosis, whereas inhibition of autophagy or overexpression of Bad and Bim reversed HCC cell resistance to mitomycin [39]. Other studies have shown that Beclin 1, a critical component of the class III PI3 kinase complex (PI3KC3) that induces autophagosome formation, also regulates apoptosis. Beclin 1 contains a BH3 domain that can bind to multiple antiapoptotic Bcl-2 family proteins, such as Bcl-2, Bcl-xL/Bcl-2L1 and Mcl-1, which may have a direct role in proapoptotic and antiapoptotic [40, 41]. Activation of autophagy by chemotherapeutic drugs leads to release of Beclin 1 from Beclin 1-Bcl-2 and Beclin 1-Bcl-xL complexes [42], further increasing autophagic flux and suppressing apoptosis due to liberation of Bcl-2 and Bcl-xL.

In conclusion, autophagy plays an important role in the resistance of HCC cells to chemotherapeutic drugs. Moreover, varying levels of autophagic flux induced by different chemotherapeutic drugs can lead to completely differ-

ent outcomes. Collectively, these studies strongly suggest that targeting the molecules involved in autophagy enhances the sensitivity of HCC to chemotherapeutic drugs.

Lysosome and transcription factor EB (TFEB)

Autophagy can be divided into four processes: induction, autophagosome formation, autophagolysosome formation, and delivery and degradation by lysosome [43]. In these processes, lysosome plays an important role as autophagy dependent organelle. Moreover, lysosome is also associated with another drug-resistance mechanism, called lysosomal sequestration or lysosomal drug entrapment. Hydrophobic weak base chemotherapeutics such as doxorubicin and vincristine accumulate in lysosomes due to the highly acidic luminal pH, which prevents these agents from reaching their intracellular targets [44]. The lysosomal lumen alkalinizers CQ and hydroxychloroquine (HCQ) are currently the only clinically available drugs that inhibit autophagy. These drugs deacidify the lysosome and block the fusion of autophagosomes with lysosomes [45]. Many studies have found that CQ can inhibit autophagy in HCC cells in vitro and in vivo, thereby enhancing tumor cell sensitivity to various chemotherapeutic agents [29-31, 46-48]. Ding et al found that oxaliplatin treatment of Huh7 and SMMC-7721 cells induced autophagy, which in turn scavenged oxaliplatin-induced reactive oxygen species (ROS) and reduced cytotoxicity, whereas the inhibition of autophagy by chloroquine enhanced ROS accumulation and oxaliplatin-induced cell death [46]. Similarly, Gao et al found that CQ inhibited autophagy and increased apoptosis induced by TACE, resulting in decreased tumor volume in the rabbit VX2 liver tumor model [48].

In the process of autophagy, the formation of autolysosome by the fusion of autophagosome and lysosome requires the depletion of large amounts of lysosomes. In order to maintain the autophagic flux of the tumor cells, tumor cells need ensure the quantity and function of lysosome by lysosomal biogenesis, autophagic lysosomal reformation (ALR) etc., thereby maintain the dynamic balance between lysosome and autophagic body, and ensure the progress of autophagy. The generation of lysosomes requires the coordinated induction of many

genes, there are more than 50 soluble acid hydrolases that perform the digestive functions of lysosomes and over 120 lysosomal membrane proteins required to maintain structural and functional integrity, including proteins that induce low luminal pH (4.5-5.0) [49, 50]. Transcription factor EB (TFEB), a member of the basic helix-loop-helix leucine zipper (bHLH-Zip) family, acts as the master regulator of lysosomal biogenesis by binding to Coordinated Lysosomal Expression and Regulation (CLEAR) motifs of genes encoding lysosomal hydrolases and membrane proteins [51]. The activity and subcellular localization of TFEB are mainly determined by the phosphorylation state of two serine residues, Ser142 and Ser211. When both residues are phosphorylated, TFEB is inactive and localized in cytoplasm. When Ser142 and Ser211 are dephosphorylated, TFEB is activated and translocated to the nucleus, where it binds to CLEAR elements promoting the expression of the entire network of lysosomal genes [52], thereby initiating lysosome biogenesis [53]. In addition to regulating lysosomal biogenesis, activated TFEB can drive the expression of autophagy-related genes and promote the formation of autophagosomes and fusion of autophagosomes with lysosomes, increasing autophagic flux [54]. Fang et al found that treatment of human colon cancer cells with doxorubicin-induced autophagy and TFEB activation. In both human colon cancer cells and human cervical cancer cells, overexpression of TFEB decreased doxorubicin-induced cell death concomitant with increased autophagic flux, whereas a small interfering RNA against TFEB blocked doxorubicin-induced autophagy and significantly enhanced its cytotoxicity. TFEB is also believed to regulate lysosomal drug entrapment [55, 56]. Zhitomirsky et al found that non-cytotoxic nanomolar concentrations of doxorubicin and mitoxantrone triggered nuclear translocation of TFEB in tumor cells, leading to a marked increase in the number of lysosomes per cell, enhancing lysosomal drug entrapment and multidrug resistance (MDR) [57]. However, few studies focus on the relation between TFEB activation and chemotherapy drug resistance in hepatocellular carcinoma. Therefore, TFEB-controlled lysosomal biogenesis and autophagy may play an important role in chemotherapy drug resistance in HCC and is a potential therapeutic target

Chemosensitization of HCC by targeting autophagy

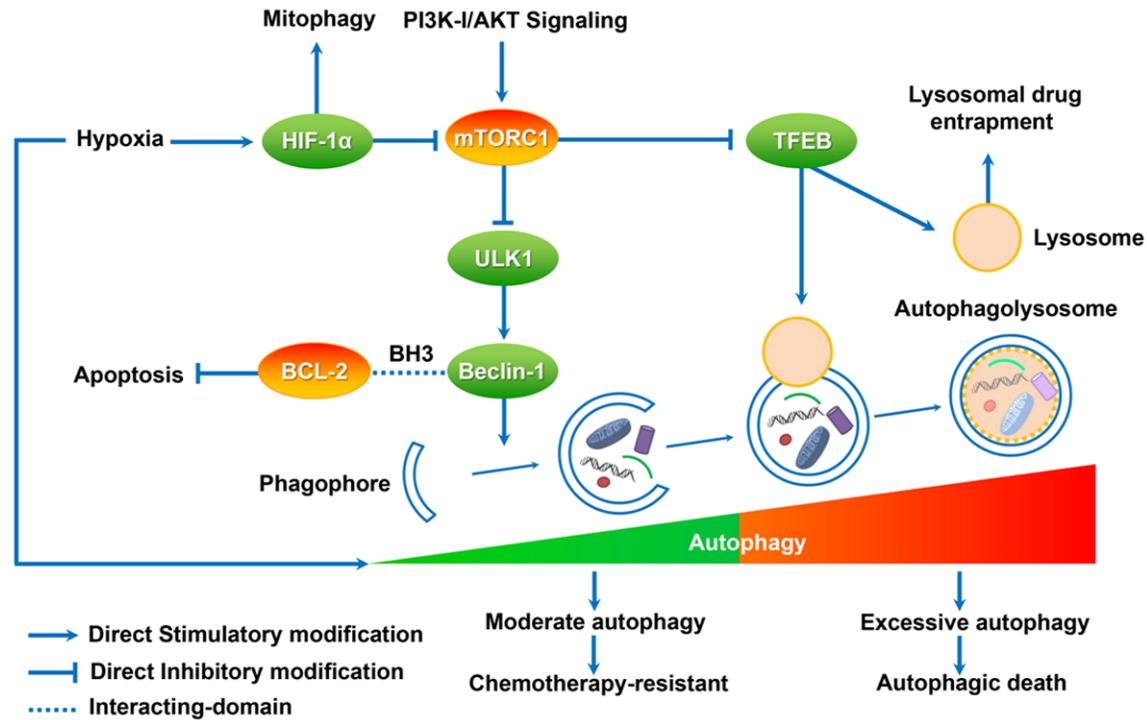


Figure 1. Potential targets of the autophagic pathway and related pathways in chemotherapy-resistant of HCC. Autophagic promoters and suppressors of are labeled in green and orange, respectively. Moderate autophagy is involved in chemotherapy-resistant of HCC, but excessive autophagy cause autophagic death. HIF-1 α , Hypoxia-inducible factor-1-alpha, mTORC1, mammalian target of rapamycin complex 1, TFEB, Transcription factor EB, BCL-2, B-cell lymphoma 2, PI3K-I, Phosphatidylinositol 3-kinase Class I.

for increasing the sensitivity of HCC to chemotherapeutic drugs.

PI3K/AKT/mTOR pathway

The phosphatidylinositol 3-kinase (PI3K)/Akt/mammalian target of rapamycin (mTOR) pathway is the most important signaling cascade controlling lysosomal homeostasis and autophagy and is hyperactivated in half of HCC cases [58]. In addition to autophagy, the PI3K/Akt/mTOR cascade is a central regulator of cell growth, survival, metabolism, and apoptosis through various downstream substrates [59]. mTOR complex 1 (mTORC1), comprising mTOR, mammalian lethal with Sec13 protein 8 (mLST8), regulatory protein associated with mTOR (Raptor), proline-rich Akt substrate of 40 kDa (PRAS40), and DEP-domain-containing mTOR-interacting protein (Deptor) [60], is the major effector of this pathway, and abnormal mTORC1 activation results in the rapid proliferation of HCC cells. The two best characterized substrates of mTORC1 are the ribosomal S6 kinases (S6Ks) and the 4E-BP family of eukaryotic

translation initiation factors, which together control multiple processes related to HCC growth, survival, and proliferation [61]. It is generally believed that the localization of mTORC1 on lysosomes is a prerequisite for its activation [62, 63]. When mTORC1 is activated, it can inhibit ULK1 complex, a key autophagy molecule, which in turn suppresses the formation of autophagosome precursors (phagophores), ultimately blocking autophagy [64]. Activated mTOR can also phosphorylate TFEB Ser142 and Ser211, leading to cytoplasmic localization and suppression of genes required for lysosomal biogenesis [63]. When tumor cells are nutrient starved or treated with mTORC1 inhibitors such as Torin1 and pp242, mTORC1 is separated from the lysosomal membrane. This free state relieves ULK and autophagy inhibition and activates TFEB-induced lysosomal biogenesis, ensuring an adequate quantity of lysosomes for autophagy [65], the induction of autophagy by sorafenib is related to the inhibition of mTOR signaling [66]. Excessive inhibition of mTOR leading to a high autophagic flux is also involved in autophagic

Chemosensitization of HCC by targeting autophagy

cell death (type-II programmed cell death) [67, 68]. Therefore, mTORC1 controls autophagic flux by regulating both the formation of autophagosomes and lysosomal biogenesis, and then controlling autophagic flux. The most important function of mTOR in tumors, however, is as a master regulator of cellular metabolism and proliferation. Therefore, mTOR inhibition can produce multiple effects including inhibition of tumor cell growth and activation of autophagy in varying degrees, it will also affect the sensitivity of HCC to chemotherapeutic drugs. Studies in vitro have found that the mTORC1 inhibitor rapamycin, a mTORC1 inhibitor, can induce autophagy in HCC, leading to enhanced resistance to chemotherapeutic drugs [47, 69]. Alternatively, other studies reported that mTOR inhibitors can enhance the antitumor effect of chemotherapeutic drugs on HCC [70, 71]. This may be due to the inhibition of abnormal mTOR pathway activation in HCC [72], resulting in the suppression of tumor cell proliferation and survival. Inhibition of mTOR in HCC can also cause autophagic cell death [73]. However, a recent randomized, multicenter, multinational phase II trial found no evidence that sorafenib combined with everolimus (a potent inhibitor of mTOR) has higher efficacy compared with sorafenib alone [74].

As the upstream of the mTOR pathway, PI3K and Akt are also critical regulators of autophagy, cell proliferation, migration, survival, and angiogenesis. Therefore, similar to mTOR pathway, inhibiting the PI3K/Akt pathway may produce a variety of effects on tumor cells in addition to enhancing autophagic flux in HCC cells [75, 76]. Zhai et al reported that GDC0068, a novel ATP-competitive pan-Akt inhibitor, reversed acquired resistance to sorafenib in two HCC cell lines by switching autophagy from a protective to a death-promoting process [77]. Simioni et al found that combining the novel allosteric Akt inhibitor MK-2206 can increase the doxorubicin-induced autophagy and apoptosis of HCC cells. However, MK-2206 enhanced the chemotherapeutic sensitivity of HCC mainly through the inhibition of proliferation and survival, while autophagy was still protective [78]. Therefore, although blocking the PI3K/AKT/mTOR pathway can suppress tumor growth by inhibiting proliferation, it is currently unclear whether the sensitivity of HCC to chemotherapeutic drugs can be enhanced by re-

gulating autophagy through the PI3K/AKT/mTOR pathway.

Hypoxia and hypoxia inducible factors (HIF)

It is widely accepted that most solid tumors, including HCC, contain regions of hypoxia [79], and most of therapies of HCC, like TACE and sorafenib can also aggravate hypoxia. Hypoxia in tumors leads to the activation of genes associated with angiogenesis and cell survival [80]. In addition, hypoxia can induce chemotherapeutic drug resistance and activate autophagy in HCC. Liang et al found that sorafenib-resistant HCC patients showed higher intratumoral hypoxia [81], and Song et al found that HCC cell lines were resistant to cisplatin, epirubicin, gemcitabine, and mitomycin in a hypoxic environment [84]. Furthermore, hypoxia increased autophagic flux, and inhibition of autophagy reversed the resistance of HCC to these chemotherapeutic drugs [82], indicating that hypoxia-induced autophagy protects HCC cells. These effects are mainly mediated by the transcription factor hypoxia-inducible factor 1 (HIF-1). HIF-1 can be activated under hypoxia and binds to hypoxia response elements to induce the transcription of genes involved in the hypoxic response [83]. Hypoxia-induced HIF-1 activity also negatively regulates mTOR, which can disinhibit autophagy [84]. Another study found that mitochondrial autophagy, also known as mitophagy, can be induced by hypoxia/HIF-1 [85, 86]. Both caspase-dependent apoptosis and ATP depletion-dependent necrosis are regulated by mitochondria [87]. Many chemotherapeutic agents will lead to tumor cell death through mitochondrial damage directly or indirectly. Mitophagy is a selective type of autophagy that degrades damaged mitochondria for quality control and protection of tumor cells from apoptosis [88]. This process is mainly mediated by four molecular pathways: Parkin (PARK2)/PTEN induced putative kinase 1 (PINK1), BCL2/adenovirus E1B 19 kDa protein-interacting protein 3-like (BNIP3L/NIX), BCL2/adenovirus E1B 19 kDa interacting protein 3 (BNIP3) and FUN14 domain-containing protein 1 (FUNDC1) [88, 89]. Prieto et al found melatonin could reverse hypoxia-mediated sorafenib resistance of Hep3B cells by preventing HIF-1 α synthesis to block the cytoprotective mitophagy [90]. Therefore, hypoxia-induced autophagy and HIF may be the targets

for increasing the sensitivity of HCC to chemotherapeutic drugs.

Conclusion and future direction

In conclusion, effective systemic treatment is the key to improve the prognosis of patients with HCC. However, most HCC patients are insensitive to chemotherapeutic drugs, and the few patients that are initially responsive to chemotherapy often develop drug resistance. Autophagy induction is a frequent response of HCC cells to chemotherapeutic drugs and leads to both intrinsic and acquired resistance. In addition, the aberrant tumor microenvironment, particularly low oxygen levels, can also induce autophagy. Therefore, interventions targeting key molecules of the autophagic pathway and related pathways, including HIF, PI3K/AKT/mTOR, pro-apoptotic, and anti-apoptotic pathways, may enhance the sensitivity of HCC cells to chemotherapeutic drugs (**Figure 1**). We believe that with the continued development of precision medicine, the role of autophagy in HCC resistance will be further clarified and that regulating autophagy to enhance the sensitivity to drugs will become an important part of individualized treatment for HCC patients.

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Disclosure of conflict of interest

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Address correspondence to: Dr. Bingjin Li, Jilin Provincial Key Laboratory on Molecular and Chemical Genetic, The Second Hospital of Jilin University, 218 Ziqiang Street, Changchun 130041, Jilin Province, China. Tel: +86-431-88796386; Fax: +86-431-88796386; E-mail: bingjinli@hotmail.com; Dr. Xuewen Zhang, Department of Hepatobiliary and Pancreatic Surgery, The Second Hospital of Jilin University, 218 Ziqiang Street, Changchun 130041, Jilin Province, China. Tel: +86-431-88796798; Fax: +86-431-88796798; E-mail: zhang-xuewen@hotmail.com

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Chemosensitization of HCC by targeting autophagy

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Chemosensitization of HCC by targeting autophagy

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Chemosensitization of HCC by targeting autophagy

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Chemosensitization of HCC by targeting autophagy

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