AUTOPHAGY AND CANCER TREATMENT REVIEW

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Abstract:

Autophagy is a catabolic process that targets impaired organelles and proteins for lysosomal degradation to maintain cell hemostasis. Autophagy in cancer is dynamic and, more specifically, depending on the stage and type of tumor. Researches in genetically engineered mouse models are agreed with the concept that autophagy can constrain initiation of the tumor by controlling oxidative stress and DNA damage, while in established tumors, autophagy can also be required for tumor survival. As shown in preclinical models, suppression of autophagy restored chemotherapy sensitivity against tumor cells. Targeting autophagy in cancer will develop a new era for anti-cancer drugs, but more specific and potent inhibitors of autophagy are needed. The role of autophagy in cancer cells continues to emerge, and further studying to identify optimum strategies to modulate autophagy for therapeutic advantage.

1. Introduction:

During the development and progression of cancer, fundamental changes in cellular processes are required to keep tumor growth. These hallmarks of cancer have been discovered during the last 30 years of cancer biology study. More recently, the autophagy process is a conserved cellular degradation pathway, which is shown to be important for various aspects of cancer biology, including protein and organelle turnover, cell metabolism, and cell survival. The autophagy role in cancer is complicated, as demonstrated by research describing situations in which autophagy can either regress or progress carcinogenesis. The most interesting explanation is that the function of autophagy in cancer is dynamic. Although autophagy constrains cancer initiation by its role in the homeostasis of the cells by maintaining genomic and cellular integrity, it is clearly required for tumor progression and may also be required for tumor maintenance, but it depends on the origin of tissue and tumor type. This review will be evaluating the role of autophagy as a target in cancer treatment and focusing on recent advances. [1,2,3]

2. Autophagy:

Macroautophagy (referred to as autophagy) is a preserved cellular catabolic process that degrades organelles and macromolecules via the lysosome to keep cellular homeostasis and

fitness, also during periods of stress. [1] Autophagy involves the coordination of more than 30 autophagy-related (Atg) proteins activity that cargo in double-membrane vesicles (autophagosomes) that fuse to lysosomes membrane (autolysosomes). Accordingly, this fusion leads to the degradation of cargo, such as damaged organelles, toxic protein aggregates, lipids, and acids, as well as pathogens, such as Salmonella (Figure 1). these breakdown byproducts of the lysosomal process (lipids, amino acids, and nucleotides) are considered as basic molecular building blocks that used in bioenergetic and anabolic pathways. [2,3]

In the beginning, autophagy is initially considered to be a nonselective, bulk degradative pathway that is stimulated in response to stressors, including starvation. However, more recent studies have shown selectivity in the autophagic pathway to identify specific cargo for degradation. [4] Although with its importance in maintaining cellular homeostasis, the disruption of autophagic function has been identified to play a part in different diseases, including atherosclerosis, neurodegeneration, andcancer. [1,2,3]
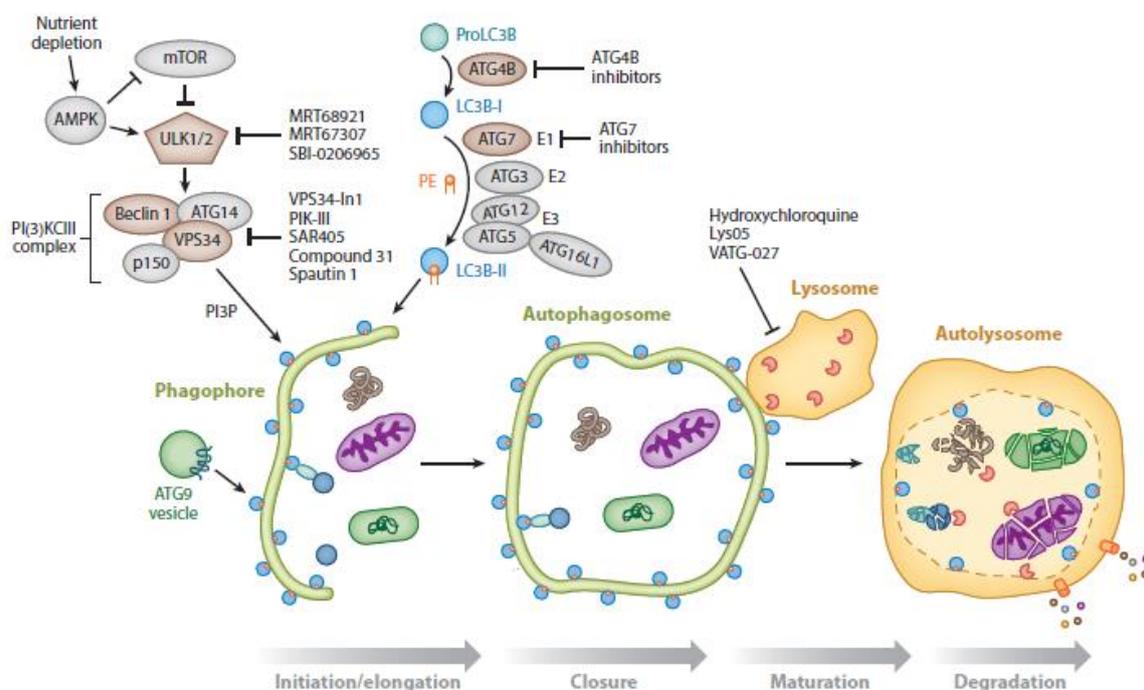

Figure2. Molecular mechanisms and the stages of autophagy (initiation/elongation, closure, maturation, and degradation) are shown. [3]

3. The molecular basis of autophagy:

3.1 An overview of the autophagy machinery

The molecular basis of autophagy is discovered by yeast genetics and then via studies in higher organisms. There are several autophagy-related genes (ATGs), up to been identified, and the ATG genes that represent the core machinery of autophagy can be categorized into many functional units: the Unc-51 like autophagy activating kinase 1 (ULK1) protein kinase complex, an initiating step for the autophagic cascade; the VPS34–beclin 1 PI3K complex; two autophagy-specific ubiquitin-like (Ubl) conjugation systems; and phosphatidylinositol-3-phosphate (PI3P) effectors and the transmembrane recycling protein ATG9. Several ATG proteins contribute to the two Ubl conjugation reactions in which LC3 and ATG12 are the

Ubl proteins. The conjugation of ATG12 to ATG5 is catalyzed by the E1-like enzyme ATG7 and E2-like enzyme ATG10. Moreover, LC3 is conjugated to phosphatidylethanolamine (PE) instead of a protein, a process that is catalyzed by ATG7, E2-like ATG3, and the ATG12-ATG5 conjugate, which is considered to be an E3-like enzyme for LC3-PE conjugation. LC3-PE conjugation (or LC3 lipidation) is essential for the formation of autophagosome, autophagic membrane binding, and cargo recognition. [5]

3.2 Molecular Mechanisms of Autophagy Initiation and Autophagosome Formation

Autophagy is initially described in mammalian tissues as an adaptive starvation response. In contrast, it is now obvious that autophagy is a vital process at many basal levels in all cells and can be more activated by different stressors, including chemotherapeutics, hypoxia, reactive oxygen species (ROS), and radiotherapy. The autophagy process can be classified into several major steps: (1) initiation and nucleation of the pre-autophagosome membrane (phagophore), (2) autophagosome closure, (3) maturation via autophagosome-lysosome fusion, and (4) degradation via lysosomal enzymes, as it is shown in (Figure 1). [5]

The main autophagy initiation pathway is controlled by multi-signaling complexes, including those that involve in cellular energy or oxidation levels (AMPK) 5_ AMP-activated protein kinase and nutrient or amino acid levels (mTOR) mammalian target of rapamycin.[6] Consequently, these pathways converge on the ULK1 complex (unc-51-like autophagy activating kinase 1, Atg1 ortholog), mediates autophagy induction. noncanonical pathways of autophagy activation not involving ULK1 or another core.[7] autophagy machinery that reflects the various mechanisms by which the autophagy can be activated. After autophagy induction, the PI (3) KCIII complex (class III phosphatidylinositol 3-kinase), including VPS34,ATG14, p150, and Beclin 1 [BECN1 (Atg6 ortholog)],autophagosome nucleates formation (Figure 1). [5]

Subsequently, the transmembrane protein ATG9 mediates the trafficking of source membrane—including from the endoplasmic reticulum, mitochondria, Golgi complex, endosome, and plasma membrane—for the autophagosome elongation stage. Two ubiquitin-like conjugation systems contribute in the step of autophagosome closure, maturation, and the recruitment of another autophagy machinery. [5,8]

3.3 Role of ATG8s in Autophagosome Maturation and Selective Autophagy

The initial component of the autophagosome maturation apparatus is the ubiquitin-like protein

lipidation system that binds phosphatidylethanolamine to the C terminus of ATG8. As a result, the growing autophagosomal membranes are incorporated with ATG8 proteins. Then, ATG7 acts as an E1 enzyme and ATG10 as an E2 to conjugate ATG5 to the ubiquitin-like ATG12 protein. Consequently, ATG12–ATG5 conjugate acts as an E3-like complex with ATG16L1 to induce ATG8 lipidation. ATG8s are manufactured in a pro-ATG8 form that is cleaved by ATG4B, leaving a C-terminal glycine residue. Finally, ATG7 (E1), ATG3 (E2), and the ATG12–ATG5–ATG16L1 (E3) complex catalyze the conjugation of phosphatidylethanolamine to the C-terminal glycine of ATG8s, then, the lipidated form of ATG8 is associated with autophagosomal membranes (Figure 1). [5]

Several studies have indicated that ATG8 proteins can act as adaptors to recruit further regulatory proteins substantial for autophagosomal maturation and as a selective autophagy receptor adaptor that physically associate their cargo to the formed autophagosomal membrane for lysosomal degradation. autophagic Selective pathways are generally named for the cargo destined for degradation and include mitophagy (mitochondria), ferritinophagy

(ferritin), aggrephagy (protein aggregates), ER-phagy (endoplasmic reticulum), and xenophagy (pathogens, including bacteria), among many. [3,4]

4. Autophagy inCancer:

The role of autophagy in cancer is too complicated, as explained by a growing study describing situations due to autophagy can either promote or inhibit tumorigenesis. The most likely explanation is that the role of autophagy in cancer is dynamic with both pro-tumorigenic roles and tumor-suppressive, which are based on multiple factors, including cellular context, tumor stage, and tissue of origin. Autophagy is implicated in numerous human diseases, particularly cancers. On the one hand, autophagy could be a tumor-suppressive mechanism. For instance, mice with heterozygotic deletion of beclin 1 are susceptible to tumorigenesis in multiple tissues, and liver-specific deletion of ATG7 leads to produce of benign liver cancer. Mechanistically, the function of autophagy in the clearance of mutagens such as disrupted mitochondria (a source of ROS) may prevent insults to DNA and hence inhibit tumor development. [9]

The accumulation of particular protein substrates that are usually targeted for lysosomal degradation can also cause tumor suppression by autophagy, as p62, an autophagy receptor protein is demonstrated to enhance tumor development through many oncogenic signaling pathways, including mTORC1, NF-κB, as well as NRF2-dependent antioxidant signaling resulting from p62-dependent sequestration of the KEAP1 ubiquitin ligase. [9] In addition, although several frequently a pro-survival mechanism, autophagy can cause cell death under specific conditions "type II programmed cell death" or "autophagic cell death." This may also act as a tumor-suppressive function. [9]

Furthermore, autophagy enhances tumor progression after the initiation of tumor growth. During solid tumor development, a progression of tumor size produces stress on cells within central regions. Due to inadequate blood supply, these regions become hypoxic and nutrient deficient. Even as angiogenesis occurs, areas within solid tumors might still suffer from metabolic stress because of glutamine availability and low glucose due to continued hypovascularization and tumor-associated vessels' leakiness. Therefore, tumor cells can use autophagy to provide an alternative energy source for proliferation and survival. Similarly, many cancer therapeutic strategies have been shown to inhibit tumor tissue angiogenesis or block the kinase signaling that usually inhibits autophagy, thus activation of cancer cell autophagy as a survival mechanism. [3]

4.1 Autophagy as a tumor inhibitor
It has been initially thought that autophagy can be a tumor-suppression process. This concept is derived from researches that the essential autophagy gene ATG6/BECN1 is lost in 40% to 75% of human ovarian, prostate, breast cancers. Indeed, in certain studies, autophagy suppression cancer cell growth, and Becn1 heterozygous mutant mice are prone to develop lung and liver tumors or lymphomas. However, mosaic or liver-specific autophagy deficiency through deletion of the autophagy genes Atg7 or Atg5 in mice promotes benign liver tumors only. In one experiment of genetically engineered mouse models (GEMMs) for hereditary breast cancer, allelic deletion of Becn1 enhances p53 activation and decrease tumorigenesis, which is the opposite of the findings that expecting if Becn1 is play a role as a tumor suppressor. Significantly, hepatomas where autophagy genes may be mutated and where their loss of function may promote cancer. The findings that autophagy deficiency in mice results in benign hepatomas suggest that autophagy may be essential in liver to suppress tumor

initiation, but also that autophagy could be required for tumor progression from benign to malignant disease. [10]

Furthermore, deficiency of autophagy causes genome instability, oxidative stress, and activation of the DNA damage response as a known factors of cancer initiation and progression (Figure 2A). This increase in oxidative stress activates the master regulator of antioxidant defense, nuclear factor NRF2 (erythroid-2–like 2), which can stimulate tumor growth. The deficiency of autophagy in liver is also toxic that is producing chronic cell death of hepatocytes and inflammation, which are known drivers of liver cancer (Figure 2A). p62 (autophagy driver) deficiency reduces both the toxicity and tumorigenesis caused by defective autophagy; this indicated that aberrant p62 accumulation that as a result of autophagy loss is partly the cause (Figure 2A). Moreover, p62 expression enhances oxidative stress and tumor growth, and stimulation of p62 expression as a result of amplification of chromosome 5q has been implicated as a factor in the development and pathogenicity of renal cell carcinoma. In contrast, p62 deficiency inhibits tumorigenesis in GEMMs for KRASG12D-driven (mouse model) lung cancer. Unfortunately, the deregulation of p62 contributes to tumorigenesis is not known yet, but it may be associated with increased oxidative stress. p62 is also a signaling adaptor that controls many oncogenic pathways, including NRF2, mTOR, and NF-κB.[10]
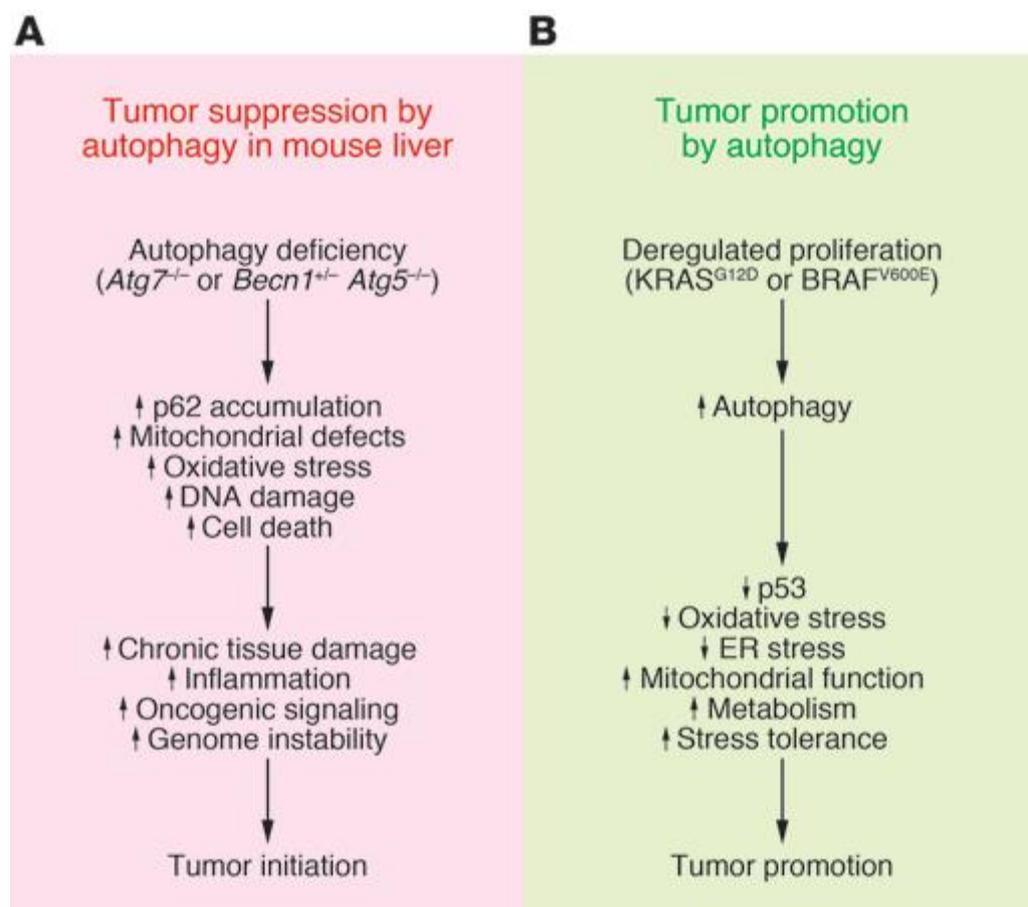

Figure 2. Proposed tumor-suppressing and tumor-promoting roles forautophagy in cancer. (**A**) Proposed mechanisms by which autophagy maysuppress tumorigenesis. Autophagy defectsimpair tissue health, leading tochronic tissue damage and regeneration that may create anenvironment thatpromotes cancer. (**B**) Proposed mechanisms by which

autophagy promotescancer by limiting stress responses and supporting metabolism and survival.[10]

4.2 Autophagy as a tumor promoter

Cancer cells mainly depend on autophagy than normal cells and tissues. Likely due to the increased metabolic and biosynthetic demands imposed by deregulated proliferation and inherent microenvironment deficiencies (Figure 2B). For example, normal autophagy is upregulated in hypoxic tumor regions, which is essential for tumor cell survival. Autophagy is upregulated in RAS-transformed cancer cells and enhances their growth, invasion, metastasis survival, and tumorigenesis. The defect in mitochondrial metabolism, which leads to an increase in the susceptibility to stress resulting from autophagy loss in RAS-driven cancers, is implicated as the underlying mechanism. These results lead to the concept that RAS-driven cancers can be "autophagy addicted." ATG17/ FIP200 deficiency suppress the development of mammary cancers in mice promoting by polyoma middle T antigen and suggesting that the role of autophagy in driving tumorigenesis and the concept of autophagy addiction in cancer can be more widely used. The genetic context that produces autophagy dependency in cancer cells is still not well understood and requires more investigation. Conversely, autochthonous GEMMs mice model for spontaneous cancers driven by the stimulation of cellular oncogenes and the loss of tumor inhibitor genes in an immune-competent host provide a more physiologic pattern address the role of autophagy in cancer cells. Given the tissue-specific, homeostatic role of autophagy, such as those with normal immune system and where tumors develop within the relevant microenvironment, is absolutely critical. [10]

Recent data show that human cancer cell lines bearing mutations in H-ras or K-ras involve a high basal level of autophagy even in the presence of nutrients. In these cells, inhibition of important autophagy protein components is shown to suppress cell growth, indicating that autophagy maintains tumor cell survival and suggests that blocking autophagy in tumors addicted to autophagy, such as Ras-driven cancers, may be an effective treatment strategy.[11]

5. Autophagy Modulation for Cancer Therapy:

5.1 Autophagy inducers

Several conventional cytotoxic therapy and irradiation have been used to induce autophagy. Other anticancer drugs that can stimulate autophagy including can be the anti– epidermal growth factor receptor (EGFR) cetuximab, BCRABL tyrosine kinase inhibitor imatinib, proteasome inhibitors, TNF-related apoptosis-inducing ligand (Table 1). The implications of enhancing autophagy in tumor cells are poorly understood and may be based on many factors, including the extent of induction, cellular context, and duration. mTOR is a central modulator of cell growth that is involving in both autophagy and protein translation. Rapamycin is a naturally allosteric mTOR inhibitor, and its analog compounds temsirolimus (CCI-779), deforolimus (AP-23573), everolimus (RAD-001) target mTORC1 selectively to induce autophagy to be effective in renal cell and neuroendocrine carcinomas and lymphoma only. Rapamycin and its analogs do not suppress mTORC2 and cannot abrogate the S6KIRS1– mediated negative feedback loop that may lead in rebound AKT activation. These limitations lead to produce ATP-competitive inhibitors for both mTORC1 and mTORC2 (e.g., Torin1, PP242, AZD8055, and WYE132) also the dual PI3K-mTOR inhibitor (NVP-BEZ235). In preclinical trials, dual inhibitors of mTORC1 and mTORC2 expressed an antitumor action and were shown to be more potent inducers of autophagy comparison to mTORC1 inhibitors alone.[11]

Table 1. Compounds that induce autophagy[12]

Compound	Mechanism and use
Rapamycin	Inhibits mTORC1
Temsirolimus/CCI-779	Inhibits mTORC1; approved for the treatment of advanced renal cell carcinoma
Everolimus	Inhibits mTORC1; approved for cancer treatment and as an immunosuppressant for organ transplant recipients
Deforolimus	Inhibits mTORC1
PP242	ATP-competitive inhibitor of mTORC1/2
Torin 1	Selective ATP-competitive inhibitor of mTORC1/2
Metformin	Activates AMPK; used to treat type 2 diabetes
CH5132799	Inhibits class I PI3K
GDC-0980	Inhibits class I PI3K and mTORC1/2
GDC-0941	Inhibits class I PI3K
Perifosine	Inhibits AKT
Fluspirilene	Reduces intracellular Ca^{2+}/calpain 1 activity and induces autophagy by preventing calpain 1-mediated cleavage of ATG5; used as an antipsychotic
Pimozide	Mechanism unknown; used as an antipsychotic
Trifluoperazine	Mechanism unknown; used as an antipsychotic
Nicardipine	Ca^{2+} channel blocker; used to treat high blood pressure and angina
Lithium	Inhibits IMPase, promotes autophagy by increasing the levels of beclin 1/VPS34 complexes
L-690330	Inhibits IMPase
Carbamazepine	Reduces intracellular inositol levels; used as an anticonvulsant and mood stabilizer
Xestospongin B	Competitive inhibitor of IP3Rs

On the other hand, the PI3K-mTOR inhibitor (NVP-BEZ235), which is in the clinical study, synergized with the use of chloroquine (CQ) to stimulate apoptosis in glioma xenografts. Another example, antidiabetic biguanide drug metformin, has been demonstrated to inhibit mTOR signaling by its upstream mediator, AMPK, and induce a cytostatic effect in certain cancer cell types. Although metformin is shown to induce autophagy in colon cancer cells, it inhibited 2- deoxyglucose–induced autophagy by reduce Beclin 1 gene expression and switch cells from survival to death in prostate cancer cells. However, this data is inconsistent with the metformin's ability to activate AMPK, which stimulates the autophagy process. Other autophagy stimulators including the selective serotonin reuptake inhibitor fluoxetine, the antiepileptic drug valproic acid, and the norepinephrine reuptake inhibitor maprotiline. In studies using cell-based screening assays, the antihypertensive drugs verapamil, clonidine, and minoxidil are shown to induce autophagy by an mTOR-independent pathway involving calpain. [11]

5.2 Autophagy inhibitors
Numerous studies have discussed that genetic knockdown of Atgs or pharmacological suppression of autophagy can effectively promote tumor cell death induced by several anticancer drugs in preclinical models, which improved response to alkylating agents in tumor cells (Table 2). In apoptosis- defective colon cancer and leukemia, suppression of autophagy is shown to re-sensitize the resistant cells to TRAIL-mediated apoptosis. Pharmacological inhibitors of autophagy can be classified as either early- or late-stage inhibitors of the pathway. Early-stage inhibitors include 3-methyadenine, LY294002, and wortmannin that target the class III PI3K (Vps34) and interfere with its recruitment to the membranes. Late-stage inhibitors include the antimalarial drugs CQ, hydroxychloroquine (HCQ), monensin, and bafilomycin A1. CQ/HCQ are lysosomotropic drugs that prevent lysosomes' acidification, whose digestive hydrolases depend on low pH, while bafilomycin A1 is a specific inhibitor of vacuolar-ATPase. Lysosomes and autophagosomes move along

microtubules, and many agents like (taxanes, vinca alkaloids, nocodazole, colchicine) inhibiting the fusion step of autophagosomes to lysosomes. [11]

Table 2. Compounds that inhibit autophagy[12]

Compound	Mechanism and use
3-MA	Inhibits class III PI3K
Wortmannin/LY294002	Inhibits class III PI3K
PT21	Inhibits VPS34 kinase
Spautin-1	Inhibits USP10/13 to promote the degradation of VPS34 complexes
Thapsigargin	A non-competitive inhibitor of SERCA that blocks the fusion of autophagosomes with lysosomes
Paclitaxel	Microtubule stabilizer that inhibits autophagy by inducing inhibitory phosphorylation of VPS34 at T159 and blocking autophagosome–lysosome fusion
SAHA	Inhibits HDACs and blocks the fusion of autophagosomes and lysosomes
CQ	Neutralizes the acidic pH of intracellular vesicles and inhibits autophagy by blocking lysosomal degradation; used to treat and prevent malaria
HCQ	A CQ derivative that blocks lysosomal degradation
Lys05	A CQ derivative with improved lysosomal accumulation
Monensin	Interferes with autophagosome-lysosome fusion; used as an antibiotic
Lucanthone	Interferes with lysosomal degradation; used as a chemotherapeutic agent and DNA intercalator
Matrine	Blocks autophagic degradation by elevating intraluminal pH of lysosomes
Xanthohumol	Prenylated chalconoid that inhibits p97 and blocks autophagosome maturation
DBeQ	Reversible ATP-competitive inhibitor of p97 that inhibits autophagosome maturation
Azithromycin	Inhibits the activity of v-ATPases and blocks autophagosome-lysosome fusion; used as an antibiotic
E64d/Pepstatin A	Inhibitors of cysteine and aspartic proteases that inhibit autophagy by blocking lysosomal degradation
Bafilomycin A1	Inhibitor of v-ATPase that blocks the lysosomal proton transport, leading to the inhibition of lysosomal hydrolases
Concanamycin A	Inhibitor of v-ATPase that blocks the lysosomal proton transport and leads to the inhibition of lysosomal hydrolases

The ability of autophagy inhibition to promote tumor regression and chemosensitivity has been confirmed in animal model studies. For instance, a Myc-induced murine lymphoma model block of autophagy by chloroquine (CQ) supported the cyclophosphamide-induced tumor cell death to an expand similar to that resulting from shRNA knockdown of Atg5 and delayed the time-to-tumor recurrence after healing. Another example, a colon cancer xenograft model; the addition of CQ to vorinostat is shown to regress tumor burden significantly and to improve apoptosis. Similarly, chloroquine combination with the Src inhibitor saracatinib enhanced the therapeutic effect of saracatinib in a prostate cancer xenograft mouse model. Also, Saracatinib decreased tumor growth by 26% as compared to control, while CQ plus saracatinib further reduced tumor growth by 64%. Interestingly, a combination also resulted in at least a 2-fold elevation in the number of apoptotic tumor cells in the group administered with saracatinib plus CQ, indicate that suppression of autophagy drives cells into apoptosis. These findings refer to autophagy inhibition to improve chemotherapeutic drugs' antitumor efficacy that follows different cellular mechanisms. Only CQ and HCQ have been estimated in humans because they are commonly prescribed in autoimmune disorders and as antimalarial drugs. Based on preclinical data, many phase I/II trials are being conducted to evaluate the combination of HCQ with cytotoxic drugs in a variety of tumor types (Table 3). [11]

Table 3. Preclinical and continuing clinical studies using the autophagy inhibitors chloroquine (CQ) and hydroxychloroquine (HCQ) in cancer treatment. [11]

Tumor type	Development status	Therapeutic combination
Colorectal cancer	*In vitro, in vivo*	CQ + bortezomib (63)
		CQ + vorinostat (56)
	Phase II	HCQ + XELOX + bevacizumab
Gastrointestinal stromal tumor	*In vitro, in vivo*	CQ + imatinib (75)
Prostate cancer	*In vitro, in vivo*	CQ + Src kinase inhibitors (57)
Vulvar cancer	*In vitro*	CQ + cetuximab (34)
Chronic myelogenous leukemia	*In vitro*	CQ + vorinostat (73)
	Phase II	HCQ + imatinib
Lymphoma	*In vivo*	CQ + cyclophosphamide (27)
Pancreatic cancer	Phase II	HCQ only
	Phase I/II	HCQ + gemcitabine
Prostate cancer	Phase II	HCQ + docetaxel
Lung cancer	Phase II	HCQ + erlotinib
Glioblastoma multiforme	Phase I/II	HCQ + temozolomide + radiation
Multiple myeloma	Phase I/II	HCQ + bortezomib
Renal cell carcinoma	Phase I	HCQ only
Breast cancer	Phase II	HCQ only
Chronic lymphocytic leukemia	Phase II	HCQ only
Advanced solid tumor	Phase I	HCQ + sirolimus or vorinostat
	Phase I	HCQ + temsirolimus
	Phase I	HCQ + sunitinib
	Phase I	HCQ + temozolomide

6. Concluding perspectives and future directions:

Autophagy acts as a dual role as a mechanism of tumor inhibitor by adapting oxidative stress in tumor cells that maintain their survival in response to a hypoxic microenvironment, increasing metabolic demands, or cancer therapy. Moreover, surviving tumor cells by the autophagy process can enhance the development of established tumors. Several preclinical findings indicate that stress-induced autophagy in tumor cells has a cytoprotective effect; therefore, inhibition of autophagy can promote cell death by different anticancer therapies. According to these mechanisms and study findings, suppression of autophagy may pave the way for a novel therapeutic. Although many drugs can inhibit autophagy, numerous of these drugs lack antitumor activity and specificity. For example, chloroquine is one of the most tested drugs in preclinical models, and other ongoing phases I and II clinical trials are evaluating hydroxychloroquine alone or in combination with cytotoxic chemotherapy, mostly in solid tumors. [3]

On the other hand, in most cells, autophagy normally occurs to confer stress resistance and maintain cellular survival under various conditions as an important cytoprotective response. Mutations in the autophagic machinery components are associated with a number of human disorders, and malfunction of autophagy is implicated in many pathophysiologies such as cardiomyopathies, Crohn's disease, infectious diseases, and neurodegenerative disorders, including Parkinson's diseases, Alzheimer's, and Huntington's. However, induction of autophagy has been discussed to play an essential role in enhancing cancer cell growth in vivo and promoting the chemotherapies resistance and metabolic changes to maintain tumor cell survival under stress and enhance metastasis. [12, 13]

To conclude, it is challenging to decide whether autophagy can generally be druggable for cancer uses without considering all the mentioned factors above. Also, it is more logical, according to preclinical studies, to focus on the inhibition effect of autophagy to cure established tumors than to induce autophagy that promotes cancer survival. However, induction of autophagy can enhance normal hemostasis and may prevent the initiation of

cancer rather than treat the already established tumor. Importantly, specific and selective molecules for inhibition or induction to modulate the autophagy pathway are the critical point after comprehensively discovering and studying all autophagy components involved in the process. Finally, depending on the experiment's findings, it looks like quite a promising approach to modulate the autophagy process to produce anticancer therapy.

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