Interaction of autophagy with microRNAs and their potential therapeutic implications in human cancers

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Abstract

Autophagy is a tightly regulated intracellular self-digestive process involving the lysosomal degradation of cytoplasmic organelles and proteins. A number of studies have shown that autophagy is dysregulated in cancer initiation and progression, or cancer cells under various stress conditions. As a catabolic pathway conserved among eukaryotes, autophagy is regulated by the autophagy related genes and pathways. MicroRNAs (miRNAs) are small, non-coding endogenous RNAs that may regulate almost every cellular process including autophagy. And autophagy is also involved in the regulation of miRNAs expression and homeostasis. Here we reviewed some literatures on the interaction of miRNAs with autophagy and the application of miRNAs-mediated autophagic networks as a promising target in pre-clinical cancer models. Furthermore, strategies of miRNAs delivery for miRNAs-based anti-cancer therapy will also be summarized and discussed.

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Introduction

The word autophagy, from Greek ‘auto’ (self) and ‘phagy’ (eating), refers to a series of conserved cellular processes. Cells digest their own cellular contents by lysosomal degradation and recycle the ingredients to maintain cell survival through autophagy [1,2]. There are at least three types of autophagy in eukaryotic cells, macroautophagy, microautophagy and chaperone-mediated autophagy (CMA), which differ with respect to the mode of delivery to the lysosome [3]. The term ‘autophagy’ usually indicates macroautophagy, which is characterized by the engulfment of cytoplasm and organelles into double-membrane bound structures, namely autophagosomes, and delivery to lysosomes. In contrast to macroautophagy, the lysosome can directly take in cytosolic components through membrane invagination in microautophagy. While CMA is only described in mammals, substrate proteins are delivered into the lysosome by a member of the Hsp70 family of molecular chaperones in cytosol [4].

Under normal conditions, autophagy remains at a low level for the maintenance of cellular homeostasis [5]. Stressful conditions, such as glucose deprivation, hypoxia and so on, can induce autophagy [6]. During stress, autophagy exerts cytoprotective effect by degrading damaged cellular contents and recycling nutrients. However, hyperactivation of autophagy will lead to cell death called as ‘autophagic cell death’. The autophagic cell death was introduced in the 1980s to describe dying cells with increased autophagic markers [7]. Recently, the term ‘autophagic cell death’ was defined as a cell death that is mediated by autophagy, which could be suppressed by pharmacological or genetic inhibition of autophagy [8]. The molecular mechanisms and the function of autophagic cell death in physiology and pathogenesis remains unclear and needs further investigation [9].

Dysregulation of autophagy has been implicated in numerous human disease [10,11], so the tight control of autophagy is very important. Autophagy is regulated by a complex network that consists of different signaling pathways and autophagy-related genes (ATGs) [12]. Recent studies have described that a lot of microRNAs (miRNAs) are involved in regulation of autophagic process. MiRNAs are a class of small non-coding RNAs that negatively regulate gene expression in transcriptional or post-transcriptional level. MiRNAs can bind to the 3’ untranslated region (UTR) of target messenger RNAs.
Regulation of autophagy by miRNAs

Zhu et al. [16] first reported that miR-30a could negatively regulate autophagic activity. Dual luciferase reporter assay indicated that miR-30a could bind to the 3'-UTR of Beclin-1 and downregulate Beclin-1 expression. Furthermore, transfection of miR-30a mimics can inhibit rapamycin-induced autophagy in T88G cells. Recently, more miRNAs have been demonstrated to regulate some ATGs and their regulators at different stages of autophagy: induction, vesicle nucleation, vesicle elongation, retrieval, and fusion.

First, autophagy induction is initiated by activation of the ULK complex, which includes ULK1/2, ATG13, FIP200 and ATG101 [17,18]. The ULK1 protein kinase is a key initiator of the autophagic process. mTOR can phosphorylate ULK1 and mammalian ATG13 (mATG13) at nutrient rich conditions, which inhibits ULK1 kinase activity. While under starvation conditions, inactive mTOR allows ULK1 to phosphorylate itself, mATG13 and FIP200, leading to further recruitment under starvation conditions, inactive mTOR allows ULK1 to phosphorylate itself, mATG13 and FIP200, leading to further recruitment of ATG complexes, such as the class III phosphatidylinositol 3-kinase (PI3KIII), to initiate autophagy. In melanoma cells, miR-290–295 cluster could inhibit expression of ULK1, and ATG7, then suppress autophagic cell death induced by glucose starvation [19]. Leucine deprivation downregulated miR-20a and miR-106b expression via suppression of their transcription factor c-Myc. Transfection with miR-20a or miR-106b mimic can inhibit leucine deprivation induced autophagy in C2C12 myoblasts. And the mechanistic study validated that miR-20a and miR-106b can directly target ULK1 and suppress its expression [20].

Isolequiritigenin, a simple chalcone-type flavonoid derived from liquorice compounds, induced chemosensitization, cell cycle arrest and autophagy in multi-drug resistant MCF7 cells. The mechanistic study revealed that miR-25 is the main target of isolequiritigenin, and miR-25 inhibition led to autophagic cell death by directly increasing ULK1 expression [21]. A recent study has reported that ULK2, another upstream autophagy initiator, is a direct target of miR-885–3p [22], so miR-885–3p might also contribute to the regulation of autophagy.

Second, vesicle nucleation is initiated by activation of the class III PI3K/Beclin-1 complex. Numerous binding partners of this complex include Bax-interacting factor-1 (BIF-1), hVPS34, ATG14L, UV irradiation resistance-associated gene (UVRAG), Rubicon, and so on. miRNA-30a/b, miRNA-376b, miR-216a, and miR-17–5p can inhibit Beclin-1 expression, thereby suppressing vesicle nucleation [23–26]. Huang et al. [27] found that Beclin-1 can also be targeted by miR-519a. Additionally, they reported that miR-630 and miR-374a can inhibit UVRAG, which interacts with Beclin-1, then lead to activation of autophagy. ATG14, a critical component of the class III PI3K/Beclin-1 complex for the nucleation of the autophagosomal membrane, was identified as the target of miR-195 [28]. RAB5A, a small GTPase, can induce autophagosome formation by its interaction with hVPS34 and Beclin-1. miRNA-101 can target RAB5A to inhibit autophagy, indicating that miR-101 modulates autophagy at the step of vesicle nucleation [29,30].

Third, vesicle elongation requires two ubiquitin-like conjugation systems: the ATG12–ATG5–ATG16L system and the ATG8–phosphatidylethanolamine system. The ATG3, ATG4, ATG5, ATG7, ATG10, ATG12, ATG16L, and microtubule-associated protein 1 light chain 3 (LC3) are involved in this phase. miR-101 and miR-376b can negatively regulate the expression of ATG4C and ATG4D [24,29]. miR-376b had the same seed sequence and consistency targets with miR-376b such as ATG4C and Beclin-1 [31]. miR-375 inhibited LC3-1 to LC3-II conversion in hepatocellular carcinoma (HCC) cells by targeting ATG7 [32]. miR-17 can reduce ATG7 expression in glioblastoma cell lines [33]. RAB5A has also been involved in ATG5–ATG12 conjugation [29]. Therefore, miR-101 may affect both at the stage of vesicle nucleation and elongation by targeting RAB5A. miR-204 can regulate autophagy in renal clear cell carcinoma (RCC) through regulation of LC3B [34]. miR-106b, miR-93, and miR142-3p modulate autophagy via targeting ATG16L [35,36]. While miR-30a/c, miR-130a, miR-519a, miR-181a, miR-374a, miR-885–3p, and miR-630 can suppress autophagy by targeting ATG5–ATG12 conjugation [27,37,38].

Finally, the process of retrieval and fusion is regulated by ATG2, ATG9, UVRAG and ATG18. A lot of miRNAs are involved in this late stage of autophagy. ATG2B was identified as a direct target of miR-130a [39]. In mammalian cells, miR-34 represses autophagy by reducing the expression of ATG9 [40]. Jegga et al. [41] analyzed transcriptional and miRNAs-mediated post-transcriptional regulation of ATGs. They found that miR-130, 98, 124, 204 and 142 are involved in regulation of autophagy-lysosomal pathway genes. UVRAG is an important molecule in the fusion process. Therefore, miRNAs targeting UVRAG such as miR-630 and miR-374a, may be involved in the regulation of the autophagosome–lysosome fusion process [27].

Besides the miRNAs and their targets described above, there are also other miRNAs involved in autophagy regulation. Apoptosis repressor with caspase recruit domain (ARC) is identified to be the anti-autophagy protein. ARC knockout mice exhibit increased autophagic activity, while ARC transgenic mice exhibit decreased autophagy activity. miR-325 could negatively regulate the translational activity of ARC, indicating ARC is a target of miR-325 [42]. The suppression of ARC by miR-325 could enhance autophagic activity in mice model.

Immunity-related GTPase family M gene (IRGM) can regulate the innate immune response by modulating autophagy. Bioinformatics analysis revealed the IRGM is a potential target of miR-196 [43]. miR-196–based regulation of IRGM affects the efficacy of autophagy in patients with Crohn’s disease [44].

Histone acetyltransferases (HATs) and deacetylases (HDACs) play important roles in protein acetylation. The miR-206 and miR-9 could regulate the expression of HDAC and HAT in Waldenstrom macroglobulinemia (WM) cells and result in autophagy-dependent cellular toxicity [45].

BCL-2 can bind to Beclin-1 and inhibit Beclin-1–dependent autophagy. Therefore, miR-182, miR-34a, miR-210, miR-205 and miR-21 [46–50], via targeting BCL-2, probably regulate autophagy through BCL-2/Beclin-1–PI3KIII pathway. The protein of p62, also known as sequestosome 1 (SQSTM1), is one of the selective substrates for autophagy, as well as a scaffold in autophagosomes. The miR-17/20/93/106 shares the same AAGUC ‘seed’ region and direct regulates p62 expression [51], indicating their potential roles in autophagy regulation. miR-155 induced by hypoxia can enhance autophagy by targeting multiple genes in mTOR signaling, including RHEB, RICTOR, and RPS6KB2 [52]. miR-100 can promote autophagy in hepatocellular carcinoma cells by targeting mTOR and IGF-1R [53].

In summary, the reported miRNAs involved in autophagy are listed in Table 1 and illustrated in Supplementary Fig. S1.
demonstrated that dependent modulation of miRNAs promotes tumor initiation and progression. Low autophagic activity may play an important role in upregulation of oncogenic miRNAs and downregulation of tumor suppressive miRNAs. This autophagy-dependent modulation of miRNAs promotes tumor initiation and progression.

### Table 1: miRNAs involved in autophagy regulation.

<table>
<thead>
<tr>
<th>miRNA</th>
<th>Target gene</th>
<th>References</th>
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<tbody>
<tr>
<td>Autophagy induction</td>
<td></td>
<td></td>
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<tr>
<td>miR-290–295, miR-20a, miR-166b, and miR-25</td>
<td>ULK1</td>
<td>[19–21]</td>
</tr>
<tr>
<td>miR-885-3p</td>
<td>ULK2</td>
<td>[22]</td>
</tr>
<tr>
<td>Vesicle nucleation</td>
<td></td>
<td></td>
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<tr>
<td>miR-30a, miR-30b, miR-376b, miR-519a, miR-17-5p, miR-376a, and miR-216a</td>
<td>BECN1</td>
<td>[16,23–27, 31,54,55]</td>
</tr>
<tr>
<td>miR-374a and miR-630</td>
<td>UVRA</td>
<td>[27]</td>
</tr>
<tr>
<td>miR-195</td>
<td>ATG14</td>
<td>[28]</td>
</tr>
<tr>
<td>miR-101</td>
<td>RAB5A</td>
<td>[29]</td>
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<tr>
<td>Vesicle elongation</td>
<td></td>
<td></td>
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<tr>
<td>miR-101, miR-376a, and miR-376b</td>
<td>ATG4</td>
<td>[24,29,31]</td>
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<tr>
<td>miR-17 and miR-375</td>
<td>ATG7</td>
<td>[32,33]</td>
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<td>miR-204</td>
<td>LC3</td>
<td>[34]</td>
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<tr>
<td>miR-519a</td>
<td>ATG10</td>
<td>[27]</td>
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<tr>
<td>miR-100b, miR-93, miR142-3p, and miR-130a</td>
<td>ATG16L1</td>
<td>[35–37]</td>
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<td>miR-181a, miR-374a, and miR-30c</td>
<td>ATG5</td>
<td>[27,37,38]</td>
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<tr>
<td>miR-630 and miR-23b</td>
<td>ATG12</td>
<td>[27,56]</td>
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<tr>
<td>Retrieval and fusion</td>
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<tr>
<td>miR-130a</td>
<td>ATG2B</td>
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<tr>
<td>miR-34</td>
<td>ATG9</td>
<td>[40]</td>
</tr>
<tr>
<td>miR-130, miR-98, miR-124, miR-204, and miR-142</td>
<td>Autophagy-lysosomal pathway genes</td>
<td>[41]</td>
</tr>
<tr>
<td>Others</td>
<td></td>
<td></td>
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<tr>
<td>miR-325</td>
<td>ARC</td>
<td>[42]</td>
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<tr>
<td>miR-196</td>
<td>IRGM</td>
<td>[43,44]</td>
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<tr>
<td>miR-206 and miR-9</td>
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<td>miR-182, miR-34a, miR-210, miR-205, and miR-21</td>
<td>BCL-2</td>
<td>[46–50]</td>
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<tr>
<td>miR-17, miR-20, miR-93, and miR-106</td>
<td>SQSTM1</td>
<td>[51]</td>
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<tr>
<td>miR-155, and miR-100</td>
<td>mTOR pathway genes</td>
<td>[52,53]</td>
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### Regulation of miRNAs homeostasis by autophagy

miRNAs generally maintain at a low level in cancer, which may be due to downregulation of important miRNA processing enzymes, such as Drosha and Dicer, in tumor cells. Inhibition of Dicer by siRNA reduced LC3-I and LC3-II expression regardless of presence or absence of the lysosomal inhibitor bafilomycin A, indicating autophagy and Dicer presence a feedback loop [39]. Gibbings et al. [57] first reported that the key ingredients of miRNAs biogenesis complexes, Dicer and AGO2, are selectively degraded by NDPS2-mediated autophagy. The autophagy-deficient cells showed increased AGO2 and Dicer, decreased ability of AGO-binding to miRNAs, and then decreased expression of miRNAs. They predicted that non-degradable Dicer–AGO2 complexes may suppress the activity of active Dicer–AGO2 complexes. The inactive Dicer–AGO2 complexes degraded by autophagy are important for the function of the Dicer–AGO2 complexes. NDPS2-mediated autophagy is essential for regulation of miRNA activity and homeostasis.

Autophagy is also involved in the regulation of miRNAs-mediated gene silencing in Caenorhabditis elegans [58], AIN-1, a key ingredient of miRNA-induced silencing complex (miRISC), is selectively degraded by autophagy. In addition to selective degradation of RNA-induced silencing complex (RISC) components, miRNA can also be directly degraded by autophagy. Lan et al. [59] demonstrated that autophagy is down-modulated and negatively correlated with the expression of miR-224 in hepatitis B virus (HBV)-associated HCC specimens. Mature miR-224 is specifically removed by the autophagosome-lysosome pathway. The pro-autophagy genes generally have inactive mutations during human cancer development, in line with the down-regulated level of autophagy in pre-malignant cells. Decreased autophagic activity may be involved in some oncogenic miRNAs upregulation (Fig. 1).

The interaction between autophagy and miRNA is extremely complex and poorly understood. There are still many unanswered questions. Many studies have shown that autophagy fluctuates during the initiation and development of cancers, and plays a paradoxical role in different stages of this malignant disease [60–62]. Generally, autophagy may function as a tumor-suppressive mechanism in the tumor initial stage, so it is often downregulated through inactivating mutations in pro-autophagy genes. However, elevated autophagy has been observed in many cancer cells under stressed conditions, such as hypoxia and anti-cancer therapy, suggesting that autophagy may serve as a cytoprotective mechanism in tumor development stage. The molecular mechanisms of how cancer cells regulate autophagy accurately for continued growth and survival are still unclear. When faced with various stresses, cells can reprogram their gene expression through complicated miRNA-involved signaling pathways. We can hypothesize that miRNAs may contribute to fluctuations of autophagy in tumor cells. For example, recent studies discovered that miR-155 is upregulated by hypoxia induced autophagy through dysregulation of mTOR pathway in human nasopharyngeal and cervical cancer cells [52]. The complicated interaction between autophagy and miRNAs needs further investigation.

### Therapeutic potential of miRNAs-mediated autophagic pathway

Autophagy has been associated with both cell survival and cell death, but the contribution of autophagy to cancer cell death remains controversial [63]. Increasing evidence indicate that autophagy is induced by the multiple anti-cancer therapeutics, including

![Fig. 1. A hypothetical schematic model of autophagy-regulated miRNA expression in tumor development and progression. Decreased level of autophagy occurs during tumor development. Low autophagic activity may play an important role in upregulation of oncogenic miRNAs and downregulation of tumor suppressive miRNAs. This autophagy-dependent modulation of miRNAs promotes tumor initiation and progression.](Image For Fig. 1)
chemotherapy, radiotherapy, and targeted therapy. Most of these anti-cancer therapeutics activate autophagy to protect cells from stress-induced damage, thereby promote drug resistance in cancer cells. On the other hand, emerging studies also reported that anti-cancer therapy-induced an autophagic cell death. Targeting autophagy may offer a novel and complex therapeutic strategy for cancer therapy.

The autophagy regulators RAB1B is a target of miR-502 [64]. The expression of miR-502 was inversely correlated with the expression of RAB1B in colorectal tumor tissues. Over expression of miR-502 can suppress autophagy and proliferation in colon cancer cells, indicating miR-502 is a new target for colon cancer. In hypoxic conditions, miR-375 inhibits autophagy and HCC cells growth by reducing expression of ATG7 [32]. miR-101 is a key regulator of autophagy through regulation RAB5A, ATG4C, and ATG4D. Inhibition of autophagy by miR-101 synergizes HCC cells to doxorubicin and fluorouracil treatment [65]. In another study, miR-101-mediated inhibition of autophagy can enhance cisplatin-induced apoptosis in HCC cells [66]. Thus, modulation of autophagy by miR-101 represents a new cancer therapeutic target.

Additionally, modulation of autophagy by miRNAs can reverse anti-cancer therapy resistance [54]. Through inhibition of Beclin-1-dependent autophagy, miR-30a can increase the sensitivity of imatinib in chronic myeloid leukemia (CML) cells and cisplatin in tumor cells in vitro and in vivo [55,67]. Furthermore, miR-30d, miR-205, miR-199a-5p, miR-101, and miR-885-3p mediated inhibition of autophagy can increase cisplatin sensitivity in tumor cells [22,66,68–70]. miR-101 can effectively reverse tamoxifen-induced autophagy and sensitize breast cancer cells to tamoxifen [29].

miR-21 is upregulated in radio-resistant glioblastoma cells. Inhibition of miR-21 sensitizes glioblastoma cells to radiotherapy induced apoptosis via modulation of autophagy [71]. Downregulation of miR-21 can also increase the chemoresistance of leukemia cells to etoposide or doxorubicin [72]. ATG12-mediated autophagy is a critical mechanism of radioresistance. In radioresistant pancreatic cancer cells, miR-23b is downregulated. Ectopic over-expression of miR-23b suppressed radiation-induced autophagy by direct targeting ATG12 and sensitized pancreatic cancer cells to radiotherapy [56]. ATG7, an essential autophagy related gene for vesicle elongation, is a target of miR-17. Cominccini et al. [33] reported that anti-miR-17 activated autophagy and sensitized glioblastoma cells to radiation and temozolomide therapy.

The role of miRNAs-mediated autophagy in cancer therapy is controversial. Therefore, miRNAs-mediated autophagy as a therapeutic strategy should be designed according to the specific tumor type, tumor environment and the disease context.

### miRNAs-based therapeutic strategies

There are two major approaches to target miRNA: miRNA reduction and miRNA replacement. miRNA can regulate the expression of multiple genes, so miRNAs-based therapeutic strategies can affect many cell biological processes, including proliferation, apoptosis, cell differentiation, and so on. The miRNAs-based therapeutic strategies might be more favorable than the conventional therapeutic strategies.

#### Chemicals and drugs modulate miRNA expression

Several chemicals and drugs can regulate the expression of miRNAs via targeting miRNA encoding genes, miRNA maturation or degradation process. Shum and colleagues developed a simple image-based strategy to screen for inhibitors of miRNAs [73]. The authors developed a miR-21 synthetic mimic together with an EGFP-based reporter cell line, in which the expression of EGFP is under the control of miR-21. Approximately 7000 compounds were screened and six compounds were identified as potential inhibitors of miR-21. This strategy offers a new opportunity to identify novel and specific inhibitors for other distinct miRNAs with potential use to modulate autophagy. Bose et al. [74] used a molecular beacon based method to screen effective inhibitors of miR-27a. They found that five aminoglycosides were able to antagonize miR-27a’s function from 14 aminoglycosides screened. The identified inhibitors interfered with Dicer function. However, most identified inhibitors have not been studied extensively in vivo, thus the efficacy and safety of these inhibitors need to be further investigated.

Several groups have reported that drugs treatment can alter the expression of miRNAs. Imatinib can induce expression of miR-203 resulting in growth inhibition of BCR-ABL1-positive leukemic cells [75]. All trans-retinoic acid (ATRA) can induce acute myeloid leukemia (AML) cell differentiation by regulating miR-663 expression [76]. Many of the biological effects of anti-cancer drugs may be mediated by its effect on modulation of the miRNAs’s expression. So it is theoretically reasonable to develop a series of pharmacy with the function of modulating expression of specific miRNAs, which play important roles in autophagy regulation as mentioned above.

#### Anti-miRNA oligonucleotides

Antagomirs, an antisense oligonucleotide, can directly bind to a target miRNA and block its function [77]. To increase stability and efficiency, the antagomir need to be modified with 2-O-methyl-group (OME), 2-O-methoxyethyl (MOE), or locked nucleic acid (LNA). The antagomirs modified with MOE have a higher affinity and specificity to RNAs compared with antagomirs modified with OME [78]. LNA modification can extremely increase antagomir’s stability and affinity. The LNA-modified anti-miR-122 has been shown to suppress liver-expressed miR-122 significantly and lead to long-term suppression of HCV infection in liver of chimpanzees [79]. Murphy et al. [80] reported that 8-mer LNA-modified anti-miR oligonucleotides inhibited miR-17, 20a, 106b, 93, 19a, and 19b-1 expression and prolonged the survival of mice in the murine medulloblastoma model. The major disadvantage of anti-miR oligonucleotides is its unspecificity. They may affect endogenous RNA species other than the target miRNAs.

#### miRNA Mimics

miRNA mimics are synthetic 18–22 nucleotide oligonucleotides designed to mimic native miRNAs. miRNA mimics are identical to the endogenous miRNAs and target the same miRNAs. Studies have shown that a two-stranded oligonucleotide is 100–1000 fold more effective than a single-stranded mimic [81]. It has been reported that double-stranded Let-7 mimics can suppress the growth, migration, as well as cell cycle progression of lung cancer cell lines in vitro [82]. miR-34a synthetic mimics triggered growth inhibition and apoptosis in multiple myeloma cells with downregulating canonic targets BCL-2, CDK6, and NOTCH1 in vitro and in vivo [83]. In mouse xenograft models, relevant tumor growth inhibition and survival improvement were observed in animals treated with miR-34a mimics in the absence of systemic toxicity. The limitations of miRNA mimic include potential off-target effects, susceptibility to nuclease degradation, and affect the normal functions of cells.

In summary, the miRNAs-based therapeutic strategies are illustrated in Fig. 2.

#### miRNA delivery systems

miRNA-based therapy need to have selective and accurate delivery systems to increase the therapeutic potential and reduce possible systemic toxicity.
Viral-based delivery system

Adeno-associated viruses (AAV) are often used for delivering miRNAs. The advantages of these vectors are that AAV vectors do not integrate into the genome and can be removed efficiently with minimal toxicity. AAV-based therapeutic strategies are in preclinical and clinical studies, including the use of novel tissue-specific promoters, and AAV capsid mutants and chimeras [84]. Tissue-specific promoters can be used to ensure efficient delivery to the specific organ. AAV-based miR-26a therapy inhibits cancer cell proliferation and increases apoptosis in a mouse model of liver cancer without significant systemic toxicity [85].

Lipid-based delivery system

Liposome is one of the majorly used transfection reagents. Lipid-based system have been shown to have increased cellular uptake, lifetime in circulation and ability to penetrate into the tumor [86]. Using chemically synthesized miR-34a and a lipid-based delivery vehicle efficiently inhibited tumor growth without liver or kidney toxicity and immune response in a mouse lung tumor model [87]. Lipid-based delivery of miR-107 mimics effectively decreased tumorigenicity of head and neck squamous cell carcinoma in mouse xenograft models [88]. Li et al. [89] developed a T-VISA-miR-34a plasmid, to specifically over-expression of miR-34a in breast cancer cells by an hTERT promoter. In an orthotopic mouse model of breast cancer, T-VISA-miR-34a liposomal complex can strikingly suppress tumor proliferation, prolong survival without systemic toxicity.

Nanocarriers delivery system

With nanocarriers, drugs can be encapsulated within vehicles based on similar building blocks. Incorporation of PEI into poly(l-lactide-co-glycolide) (PLGA) particles has good biocompatibility and biodegradability and can be useful in the gene delivery. Liang et al. [90] developed a PLGA/PEI nanoparticle for miRNA delivery in human hepatocellular carcinoma cells. They reported that PLGA-based gene delivering nanoparticle enhanced suppression effect of miR-26a in HepG2 cells. Minjun et al. [91] developed protamine sulphate (PS)-nanodiamond (ND) nanoparticles to deliver miRNA-203 into esophageal cancer cells. Griveau et al. [92] reported that delivering miR-21 antagomir with a lipid-based nanoparticle system enhanced radio-sensitivity in glioblastoma cells.

In summary, the advantages and disadvantages of different miRNA delivery systems are listed in Table 2. Many miRNAs with the potential function to regulate autophagy are disregulated in human cancers. Targeting these miRNAs for autophagy modulation presents a promising therapeutic strategy for cancer therapy. Using miRNA mimics or inhibitors, we can inhibit cytoprotective effect or promote cytotoxic effect of autophagy in a variety of cancers. Furthermore, modulation of autophagy by miRNAs-based therapy can also be used as an adjuvant therapy to circumvent cancer drug or radiation resistance.

Conclusions and perspectives

Autophagy is a basic catabolic process involved in cancer cell’s survival. miRNA-mediated regulation of autophagy related gene is an essential part of the molecular mechanisms in autophagy. Thus a thorough understanding of the interaction between autophagy and miRNAs is important to develop a miRNAs-based therapy. Until now, the relationship between autophagy and miRNAs is mainly investigated in tumor cells, but their interaction in normal cells remains unclear. Further investigation in normal cells will contribute to reveal the function of miRNAs-mediated autophagy in both physiological and pathological conditions.

However, there is still a long way to go before the clinical application of miRNAs-based therapeutic strategies. The main issues of this novel therapeutic method are as follows. The effect of chemical modification on miRNA mimics on its biological activity, the techniques for efficiently delivery of miRNA mimics or miRNA antagonimers in vivo, and the off-target effects of miRNAs-based therapeutic strategies. All these questions need to be further investigated.

Conflict of interest

The authors declare no conflict of interest.

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References


