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Review Article

Extracellular vesicles degradation pathway based autophagy lysosome pathway

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Abstract: As an ancient intracellular degradation pathway, the autophagy lysosome pathway exists in various cells continuously and stably and maintains cellular homeostasis by degrading damaged organelles and misfolded proteins that are prejudicial to cells. Extracellular vesicles (EVs) including microparticles and exosomes, are derived from varieties of mammalian tissue cells such as platelets, endothelial cells, cardiomyocytes. Through large quantity of active substances carried by EVs, EVs exert momentous biological functions. Recent researches have revealed the molecular mechanism of the interaction between extracellular vesicles and autophagy. In this review, we first elaborate that extracellular vesicles are identified and internalized by target cells by means of receptor-ligand. Since extracellular vesicles contain multiple functional molecules, we subsequently describe the process of intracellular autophagy pathway induced by extracellular vesicles, which activates autophagy-related pathways or delivers autophagy-associated molecules. Finally, we introduced the effects of extracellular vesicle-induced autophagy on extracellular vesicles and target cells respectively. In conclusion, this article integrates relevant theoretical knowledge of autophagy caused by extracellular vesicles and provides a new direction for the study of extracellular vesicles in the future.

Keywords: Extracellular vesicles, autophagy, exosome, microparticle, internalization, signaling pathway

Introduction

Extracellular vesicles (EVs) are verified to be ever more important to physiologic processes and pathophysiologic changes in human beings in recent decades. EVs, including apoptosis bodies, microparticles, and exosomes, which are 1-5 μm, 100-1000 nm, 30-100 nm in diameter respectively, play a crucial role in both normal and pathological conditions. However, current test methods can hardly distinguish microparticles and exosomes in an accurate way. Therefore, we refer to exosomes and microparticles as extracellular vesicles. EVs, derived from multiple types of cells, have a variety of biological functions such as procoagulation [1], proinflammatory [2], vasoconstriction [3], angiogenesis [4], endothelial dysfunction [5] and so on.

Owing to the existence of these functions, exploring the fate of EVs in the body is becoming more and more necessary. A large number of studies have reported that EVs can be internalized by various types of cells. Nevertheless, the internalization process cannot fully represent the degradation of EVs, thus further investigation of the intracellular degradation pathways after EVs entering cells is necessary.

Autophagy is the extremely important intracellular degradation pathway by which cytoplasmic cargos like misfolded proteins, aging organelles are degraded after being delivered into the lysosome [6]. Jochen Klucken et al. showed that EVs derived from multivesicular body and were eliminated by the autophagy lysosome pathway [7]. Accordingly, we hypothesize that the autophagy lysosome pathway plays a key role in the clearance of extracellular vesicles.

Autophagy

The conception of autophagy

Autophagy is a unique and conserved protein degradation pathway in cells [8], which can be

activated by various stimuli like hypoxia, ischemia, tumor and so on. In the process of autophagy, intracellular unwanted materials are equipped into the autophagosome and then are degraded in the lysosome for recycling to maintain cellular homeostasis. The normally functioned autophagy-lysosomal pathway (ALP) can transfer cellular intrinsic or foreign substances into lysosomes for degradation whereas the dysfunctional ALP loses the above degradation function [9].

The role of autophagy in physiological or pathological conditions

It is a known fact that autophagy exists in normal and pathological circumstances. However, the function of autophagy is different in different situations. In normal situations, basal autophagy has a role in inhibiting tumorigenesis by eliminating redundant oxygen free radicals and unstable genomes. Autophagy can be induced by physiological signals such as starvation, in order to facilitate cell survival [10]. Maurizio Battino et al. proposed that autophagy has been proven to be an important regulator of aging by removal of misfolded proteins, mutant α-synuclein, damaged mitochondria as well as tau which participate in the development of multiple cumulative diseases and recycling of cytoplasmic substance [11]. Therefore, moderate autophagy plays a protective role under diversified physiological situations.

Nevertheless, excessive autophagy is detrimental and results in programmed cell death [12]. An ever increasing number of researchers have linked autophagy to pathological conditions [13]. Zhang et al. demonstrated that autophagy was considerably enhanced in hypoxia diseases including OSAHS, traumatic brain injury and myocardial infarction [14]. Myocardial ischemia-reperfusion injury increased the expression of autophagy markers like LC3II/I, Atg5 and Atg7, whereas above protein changes were reversed by silencing of lncRNA AK139328, thereby protecting cardiomyocytes away from autophagy and apoptosis [15]. These findings suggest that severe autophagy aggravates the onset and progression of related diseases and ultimately leads to a poor prognosis. Hence, the regulation of intracellular autophagy is extremely meaningful.

Recognition and internalization of extracellular vesicles by target cells

Cells that recognize EVs

EVs are small vesicles that are detached from the cell membrane after cell injury, activation or apoptosis. These nano vesicles carried a variety of biologically active substances such as proteins, lipids and RNAs [16]. In recent years, the multiple effects of extracellular vesicles outside the cells have been investigated, but the role in cells is largely unclear. Many studies have shown that extracellular vesicles can be recognized by a number of cells such as endothelial cells [17], macrophages [18], dendritic cells [19], cardiomyocytes [20] and tumor cells [21]. This may be the premise that extracellular vesicles are internalized by target cells and activate intracellular biological signals.

The approach in which EVs are internalized by target cells

A 52-kDa glycoprotein called Developmental endothelial locus-1 (Del-1) is secreted by endothelium. Del-1 is expressed by endothelial cells, as well as exists in a certain subset of macrophages. Its amino terminus contains three EGF-like repeats which can combine with integrins αVβ3 and αVβ5 on endothelial cells, meanwhile its C-terminus consists of two discoidin I-like domains which enable Del-1 to bind phosphatidylserine on the surface of EVs [22]. Consequently, Del-1 is one of the most significant mediators for internalizing extracellular vesicles. As mentioned above, lactadherin, a 41-46 kDa glycoprotein secreted by macrophages, tethers PS-enriched extracellular vesicles to macrophage integrins. Namely, lactadherin also has an epidermal growth factor-like domain including a RGD sequence at N-terminal, which can combine with integrins of macrophages and two discoidin I-like domains that bind PS closely at C-terminal [23]. In summary, as a bridge between extracellular vesicles and target cells, lactadherin plays a critical role in the clearance of extracellular vesicles.

Another major discovery is annexin I/PSR dependent pathway. Nikos Werner et al. indicated that annexin I was highly expressed in microparticles derived from endothelial cells (EMP) meanwhile the PSR was expressed on

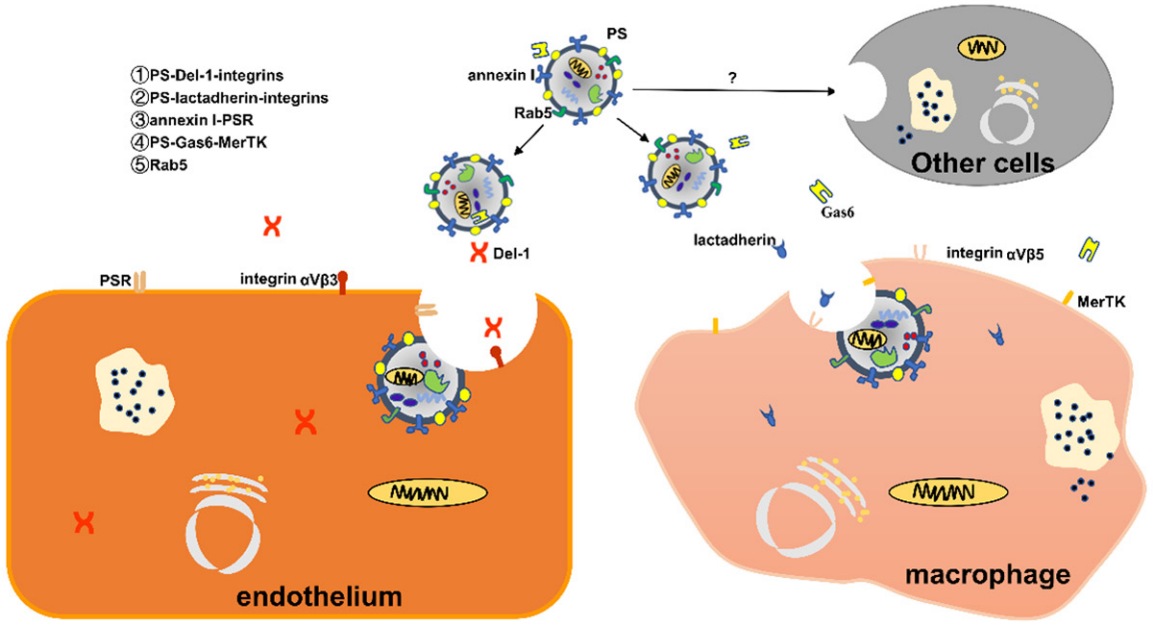

Figure 1. The approach in which EVs are internalized by target cells. This figure summarizes the fact that extracellular vesicles are recognized and internalized by target cells in a variety of receptor-dependent manners. In these ways, phosphatidylserine (PS) exposed to the surface of extracellular vesicles acts the most important role. For example, PS-Developmental endothelial locus-1 (Del-1)-integrin, PS-lactadherin-integrin and PS-Gas6-MerPK are shown in the figure. In addition, the annexin I/PSR pathway and Rab5 on the surface of extracellular vesicles also partially mediate the extracellular vesicles being internalized by target cells. Apart from these receptor-dependent ways, some previously reported endocytosis and macropinocytosis also promote uptake of extracellular vesicles.

the surface of HCAEC. Blockage of annexin I or PSR significantly inhibited EMP endocytosis by target cells [24]. These findings demonstrate that the annexin I/PSR pathway is indispensable for HCAEC to take up EVs.

TAM receptors including tyro3, Axl, and Mer were able to recognize PS on apoptosis cells [25]. Interestingly, Gas6 and protein S secreted by microparticles could act as bridge molecules between TAM and PS. Mer highly expressed by Alveolar macrophages promoted macrophages' uptake of PS-containing microparticles [26]. These evidences suggest that MerTK is a key signaling molecule in the process of macrophage uptake of microparticles (**Figure 1**).

In addition, a special protein Thy1 expressed on the surface of mesenchymal stem cells can be secreted from cells by extracellular vesicles. More importantly, Thy1 were able to interact with integrins on the EVs and mediated extracellular vesicles by fibroblast internalization [27]. Recently, studies have also found that Rab5, a GTPase that regulates endosome formation and fusion of endogenous vesicles with endosomes was in a position to mediate the internalization of microvesicles derived from erythrocyte by lung endothelial cells. After silencing Rab5 in endothelial cells, its ability to ingest microparticles was markedly reduced [28]. Furthermore, endocytosis and macropinocytosis were also engaged in the endocytosis of extracellular vesicles [29]. Of course, there are still numerous recognition modes that need further exploration.

Activation of intracellular degradation pathway by extracellular vesicles

The signal pathways

In recent decades, with the in-depth study of autophagy pathway, cellular autophagy has been understood commonly. However, the discovery of extracellular vesicles with multiple physiological or pathological effects once again arises numerous researches in the field of autophagy.

Akt/mTOR signal pathway

It is well known that autophagy is related to numerous signaling pathways, and among

these autophagic pathways, the PI3K/AKT/mTOR pathway is one of the most studied pathways, which acts as an important role in various cellular processes such as transcription, growth, proliferation, survival and angiogenesis [30]. PI3K recruited and activated downstream signaling targets such as AKT after converting phosphatidylinositol-4,5-bisphosphate to phosphatidylinositol-3,4,5-triphosphate [31]. Structurally, mTOR is a complex composed of mTORC1 and mTORC2, of which mTORC1 is a key negative regulator of autophagy [32]. Recently, in vivo experiments have found that compared with pure diabetic nephropathy (DN) mice, mesenchymal stem cell-derived exosomes significantly increased the expression of Beclin-1 and LC3-II in the kidney of DN mice through the induction of mTOR signaling pathway, which delayed the progression of DN. The renal protection of exosomes was repeatedly verified by changes in blood biochemistry and renal histology [33]. A large number of reports have already explained the importance of mTOR and its downstream molecule p70S6K in regulating autophagy [34]. As a major downstream target of PI3K/AKT, mTOR is deemed to be a key molecule regulating mammalian growth and metabolism [35]. Mitofusin2 activated autophagy process of pancreatic cancer by suppressing the PI3K/AKT/mTOR pathway [36]. Perfluoroalkyl acid exposure initiated protective autophagy through inhibiting the PI3K/AKT pathway [37]. A recent research has shown that exosomes from mesenchyme stem cells inhibited myocardial remodeling after ischemia/reperfusion injury by activating PI3K/AKT signaling pathway in vitro [38]. Shen et al. found that the level of autophagy increased significantly after MSC-derived exosomes being internalized by H9C2 cell. Simultaneously, the expression levels of p-mTOR/mTOR and p-AKT/AKT were dramatically reduced whereas the ratio of p-AMPK and AMPK was significantly increased in the H_2O_2+exosomes group compared with H_2O_2-only group. In consequence, activated intracellular autophagy by MSC-exosomes was at least partially mediated by these two autophagy pathways [39]. According to several experiments, induction of the PI3K/AKT/mTOR signaling pathway can reduce immoderate autophagy that leads to cell death and enables to promote cell survival. In addition, inhibition of mTORC1 can stimulate autophagy of harmful substances and reduce intracellular toxicity.

STAT3/BCL-2/Beclin-1 pathway

As a negative regulatory molecule of autophagy, STAT3 was previously reported to act a crucial impact on the autophagy process by reducing the activity of protein kinase R [40]. Mesenchymal stem cells suppressed the expression of STAT3 and promoted the occurrence of autophagy. However, MSCs could not augment autophagy of HiBECs treated with Glycochenodeoxycholate after silencing STAT3. This phenomenon also proved that the autophagy and STAT3 had opposite trends [41]. We all know that apoptosis and autophagy are two important processes to maintain the homeostasis of the body. A growing number of researches have proved that the crosstalk of autophagy and apoptosis is quite complicated [42]. Bcl-2, an anti-apoptotic protein, can interact with Beclin-1. The interaction between Bcl-2 and Becline-1 is a major inducer of autophagy [43]. With the gradual deepening of extracellular vesicles research, many researchers are wondering whether extracellular vesicles can activate autophagy by activating certain signaling pathways. Lu et al. showed that the expression of STAT5 and Bcl-2 in HSC-T6 cells was down-regulated by exosomes containing miR-181-5p whereas the Beclin-1 expression was up-regulated by miR-181-5p-rich exosomes. These evidence also strongly suggests that exosomes can activate autophagy through the STAT3/BCL-2/Beclin-1 pathway [44].

Toll-like receptor-ligand signal pathway

TLRs are largely expressed in a variety of tissues like renal tissue and inflammatory cells, which act as a vital constituent on the induction of autophagy. Especially TLR2, one of the TLR family members, exerted a protective effect by up regulating autophagy in acute kidney injury induced by cisplatin [45]. Another study found that the expression of TLR2 and TLR4 in hippocampus neurons of epilepsy mice was significantly up regulated. Overexpression of miR-421 in hippocampus neurons of epilepsy mice could inhibit autophagy process by inhibiting TLR/MYD88 signal pathway [46]. A previous study demonstrated that Lipoprotein contained in mycobacteria had a dazzling ability to activate autophagy of monocytes via TLR2/1/CD14 signal [47]. A recent surprising discovery by García et al. was that TLR2/6 ligands were discovered in EV-TB (extracellular vesicles released by

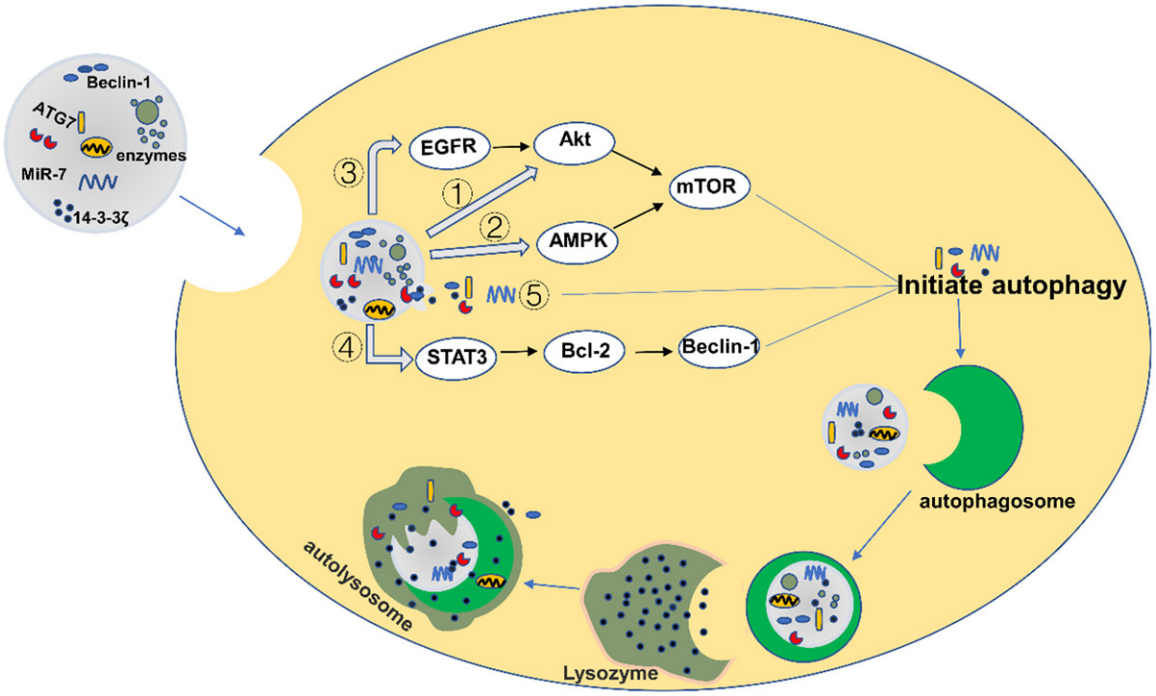

Figure 2. Activation of autophagy process by extracellular vesicles. This figure summarizes the activation of autophagy process by extracellular vesicles. After the extracellular vesicles carrying a variety of biologically active substances enter the cells, the autophagy is initiated mainly by two ways. One of them is that extracellular vesicles directly release active substances that activate autophagy, such as MiR-7, 14-3-3, Beclin-1, ATG7, etc. Another more important way is that extracellular vesicles activate multiple autophagy signaling pathways. A large number of autophagosomes form after autophagy is activated by extracellular vesicles. These autophagosomes then encapsulate harmful substances such as extracellular vesicles, multivesicular bodies, and damaged mitochondria, which are ultimately delivered to lysosomes for dissolution to maintain cell homeostasis.

human neutrophils infected with mycobacteria), moreover, the TLR2/6-rich EVs successfully activated autophagy of macrophages after internalization [48].

AMPK/mTOR signal pathway

AMP-activated protein kinase, an important cell homeostasis regulator, is indispensable for autophagy regulation. AMPK is positively correlated with autophagy while mTOR is negatively correlated with autophagy. Lee et al. proved that ezetimibe successfully activated autophagy via the AMPK-TFEB pathway [49]. Activation of AMPK significantly inhibits mTOR and activates autophagy. Research manifested that hydrogen sulfide protected ischemic myocardium by AMPK-mediated autophagy [50]. Zang's experiment showed that the phosphorylation of AMPK prominently increased whereas the mTOR phosphorylation was declined sharply in H/R myocardium treated with Ach [51]. A compelling discovery is that exosomes produced by mesenchyme stem cells induce autophagy of cardiomyocytes via AMPK/mTOR pathways to relieve myocardial ischemia/reperfusion injury [39].

EGFR/Akt/mTOR signal pathway

Studies have suggested that EFGR (epidermal growth factor receptor) is an essential molecule for many life processes such as proliferation, differentiation or survival of mammalian cells [52]. As an upstream signal of Akt/mTOR, EFGR also has an important role in autophagic regulation. Zhou et al. found in the experiment that exosomes containing miRNA were capable of inducing autophagy, which was associated with EGFR/Akt/mTOR pathways [53]. Therefore, this pathway can also serve as a bridge between extracellular vesicles and autophagy (**Figure 2**).

The active substance contained in extracellular vesicles

EVs are heterogeneous nano vesicles shed from almost all of the cell. They carry a variety

of biologically active substances, such as carbohydrates, lipids, proteins and a series of different nucleic acids including DNA, mRNA, and microRNAs [54, 55]. Autophagy of target cells is activated after the extracellular vesicles containing certain specific active molecules are internalized by cells. Therefore, we believe that bioactive molecules at least partially mediate intracellular autophagy caused by extracellular vesicles.

MiR-7

Unlike mRNA, MicroRNAs are small, short and non-coding RNAs [56]. MicroRNAs were highly conserved and first discovered in 1993 [57]. The role of microRNAs is to regulate post-transcriptional protein expression and life processes [58]. It has previously been reported that miR-7 inhibited the expression of α-Syn by binding to the 3'-untranslated region of α-synaptic protein mRNA while it could also scavenge α-Syn by activating autophagy [59]. More and more studies show that microRNA is closely related to the regulation of autophagy, such as miR-181-5p [44], miR-30d-5p [60], miR-221/222 [61], miR-30a [62] and so on. Chen et al. discovered that exosomes derived from X-ray irradiated astrocytes contained large amounts of miR-7. Moreover, miR-7-rich exosomes enhanced autophagy of lung by targeting Bcl-2 in vitro and in vivo after partial brain irradiation [63]. Another research found miR-7-5p-containing exosomes derived from human bronchial epithelial cells treated with ^{60}Co γ-rays could induce autophagy of unirradiated recipient cells [53]. In this way, extracellular vesicles can activate autophagy in target cells by transmitting specific miRNAs, which play a fateful role in a series of biological processes.

mRNA of autophagy-associated molecule

Beclin-1 is considered to be a mammalian homologous gene of the yeast ATG6 gene, which plays a decisive role in the process of autophagosome formation by interacting with Vps34 and Atg14 [57, 64]. Previous studies have suggested that beclin-1 was a component of the lipid kinase complex and was closely related to the accumulation of intracellular lipids [65]. We are also aware that autophagy is a strict intracellular degradation process that requires the biosynthesis of autophagosomes and the fusion of autophagosomes with lysosomes to degrade unwanted substances like proteins, lipids and so on. The increasing ratio of LC3-II/LC3-I generally represents an increase in autophagy [66]. A lot of research evidence has also proved that LC3-related phagocytosis is indispensable in eliminating excess substances in cells [67]. Recently, research has found that young microvesicles (MVs) derived from MSCs contained a large number of autophagy-related mRNAs such as LC3, Beclin-1 and ATG7, which enhance the autophagy of aging hematopoietic stem cells and further improve the function of hematopoietic stem cells. However, this phenomenon was reversed when RNase treatment was given, which further proved that RNA in MVs acts a major role [68]. These results provide us with a novel suggestion that extracellular vesicles may initiate autophagy through autophagy-related molecules carried by themselves after entering the target cells.

14-3-3ζ

Cell survival and death are precisely regulated by various signaling pathways [69]. The 14-3-3 protein family contains seven subtypes namely β, γ, ε, ζ, η, σ and τ, which are transcribed and translated by different genes and located in various tissue cells [70]. The 14-3-3s play a protective role in a variety of recipient cells by interacting with several signaling proteins implicated in cell regulation [71]. Among the seven subtypes that have been reported, 14-3-3ζ has the function of protecting target cells from stress damage such as ischemia and hypoxia [72]. Meanwhile, 14-3-3ζ can regulate a variety of signaling pathways related to autophagy such as PI3K and mTOR. Studies have also found that 14-3-3ζ were able to interact with ATG9A via Ser761 phosphorylation in the 14-3-3ζ binding site of the C-terminal domain of the ATG9A. In the case of normalcy, this phosphorylation was regulated by ULK1 and AMPK, thereby maintaining a low level of interaction. However, AMPK was fully activated, resulting in a markedly increase in phosphorylation of Ser761 and 14-3-3ζ binding [73]. Therefore, 14-3-3ζ is largely involved in the regulation of autophagy [69]. Qian et al. found that hucMSC-exosomes actived autophagy of recipient cells through a large number of 14-3-3ζ proteins carried by themselves. The protective effect of exosomes was greatly diminished after knock-

ing out the 14-3-3ζ gene in hucMSC or HK-2 cells. Although autophagy was not entirely blocked after gene knockout, it also indicated that 14-3-3ζ at least partially mediated the process of autophagy induced by extracellular vesicles [74].

The effect of autophagy on extracellular vesicles

Some previous experiments have found that extracellular vesicles carry a variety of substances that are detrimental to target cells. When these extracellular vesicles are internalized, a series of pathological changes occur in the target cells. For instance, high dose of microparticles derived from human umbilical vein endothelial cells exposed to hypoxia/reoxygenation conditions significantly increased the activity of caspase-3 while dramatically reducing the proportion of bcl-2/bax in H9C2 cells. In addition, the endothelial microparticles could also produce large amounts of ROS to cause apoptosis and oxidative stress of target cell [54]. Another study found that chronic myeloid leukemia-derived exosomes were in favor of tumor cell survival and proliferation through TGF-β1 receptor-ligand manner [75]. However, there are quite a few experiments that have demonstrated the potential therapeutic effects of extracellular vesicles produced by certain cells or specific stimuli. At the same time, many studies have shown that the dual role of extracellular vesicles is closely related to the level of autophagy in target cells. From the above, the autophagy of target cells caused by extracellular vesicles has an extraordinary significance for cells and the entire body.

Although the researchers are expected to explore the fate of extracellular vesicles, there are still not adequate researches. However, there is no denying that extracellular vesicles can indeed play a cytoprotective role by causing intracellular autophagy. In turn, autophagy also plays an indispensable role in the synthesis and degradation of extracellular vesicles. As one of the two major protein degradation pathways, which included the ubiquitin-proteasome pathway and the autophagolysosomal pathway, autophagosomes could fuse with multivesicles in advance and formed amphisomes before fusion with lysosomes, eventually leading to degradation of lysosomal contents such as extracellular vesicles [76]. Prior to this, the most important thing was that the extracellular vesicles were transported to lysosomes. It has been reported that extracellular components can be delivered to the degradation pathway via the endo-/exosomal pathway, while intracellular components can be delivered to lysosomes via autophagy process [77]. Therefore, extracellular components such as extracellular vesicles are very likely to be transported to lysosomes by the above two methods and subsequent degradation. It is well known that the maturation of early endosomes further leads to the formation of multivesicular bodies containing large numbers of intraluminal vesicles. These intraluminal vesicles called exosomes are released after the fusion of the multivesicular body and the plasma membrane [78]. Gorski et al. reported that autophagosomes had a strong ability to fuse with lysosomes and then cleaved their engulfed contents. Alternatively, autophagosomes were able to fuse with multivesicular bodies to form navel organelles named amphisomes, which was thought to eventually fuse with lysosomes and dissolve internal substances such as intraluminal vesicles (ILVs) [79]. The content of ILVs was significantly decreased after the multivesicular body was fused with lysosomes. In macro-autophagy, the entire cytoplasmic region was enveloped inside autophagosomes, followed by fusion of autophagosomes with endocytic vesicles (as multivesicular bodies) or lysosomes, which provided a mass of hydrolytic enzymes that would degrade the content of autophagosome [80]. When extracellular vesicles entered the cell, the cargo they carried must exit that inherent degradation pathways such as endosomes into lysosomes, or be released by MVB-plasma fusion pattern [81] (**Figure 3**). Therefore, intracellular autophagy plays a decisive role in the number and fate of extracellular vesicles produced by various stimuli. Increased levels of autophagy significantly inhibit the release of exosomes, which is due to the increased fusion of multivesicular bodies with autophagic vacuoles [82]. Previous experiments have found that exosomes from central or peripheral neurons of mice contained large amounts of prions. Up-regulating the level of autophagy in neuronal cells could inhibit the release of prions in exosomes, which in turn provided a new therapeutic target for related diseases [83]. Conversely, the inhibition of autophagy promoted the release and transfer of SNCA/α synuclein in EVs [7]. In contrast to the release of extra-

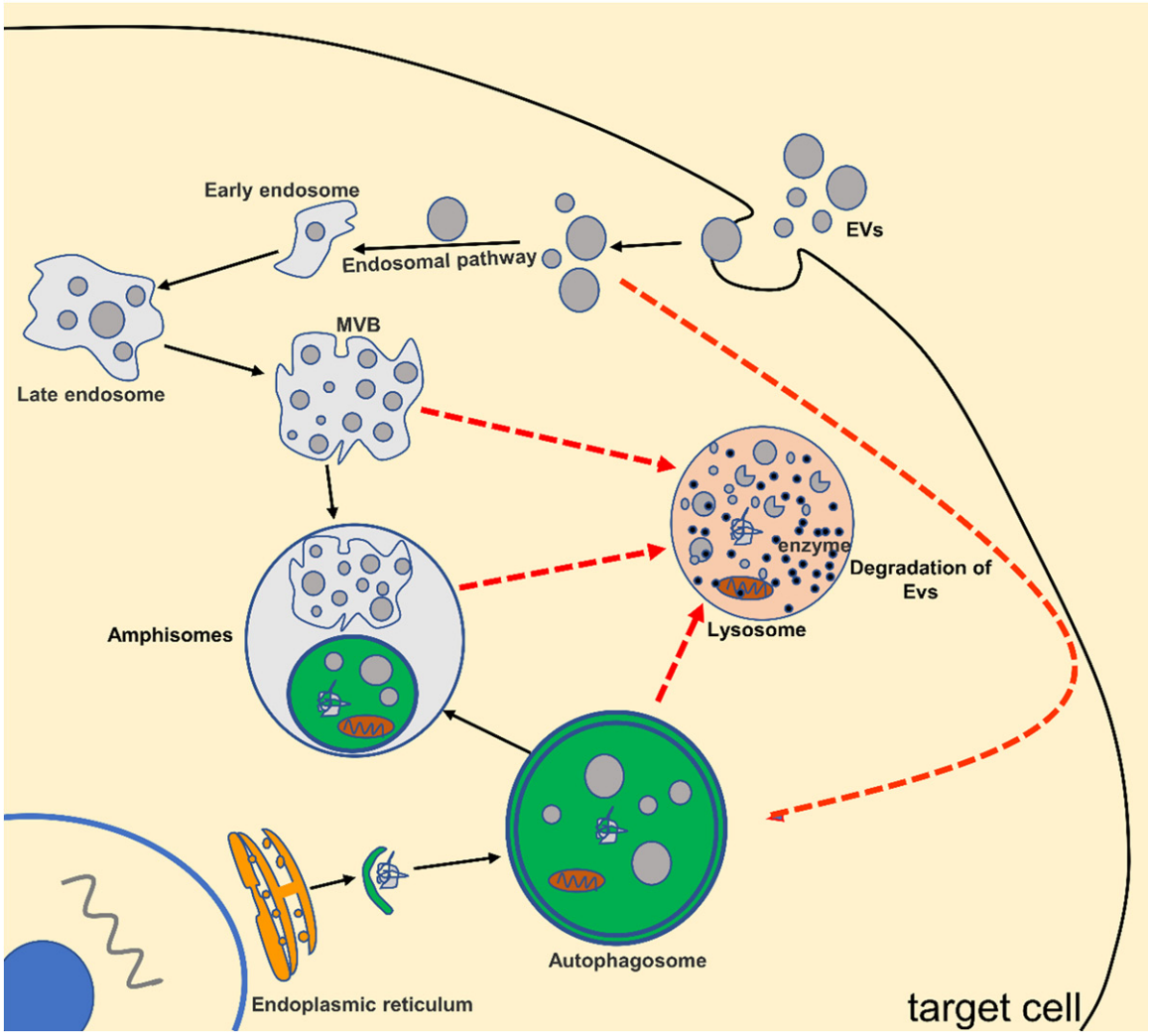

Figure 3. Degradation of extracellular vesicles by autophagy lysosome pathway. This figure summarizes degradation of extracellular vesicles by autophagy lysosome pathway. As the extracellular components, part of EVs can be delivered to the degradation pathway via the endo-/exosomal pathway. Another part of the EVs contained in the multivesicular body (MVB) is fused with the autophagosome to form amphisome, which is then delivered to lysosomal degradation. At the same time, EVs internalized by target cells can also be directly delivered to lysosomes via autophagy process for degradation. The detailed mechanism of each path in the figure requires us to further study in future work.

cellular vesicles, multivesicular body (MVB), the precursor of exosome, could be cleared by the autophagy-lysosomal pathway by direct fusion with lysosomes or autophagosomes. This is due to the fact that the multivesicular body directly enters the autophagy pathway after activation of autophagy and then significantly reduces the release of exosomes [84]. At present, although no studies have directly reported the detailed degradation process of extracellular vesicles by autophagy, some experiments have linked the degradation of extracellular vesicles and their contents to the autophagosome lysosome pathway. Therefore, the in-depth degradation mechanism of extracellular vesicles by the autophagosome pathway may become one of the future research hotspots.

At present, we have realized that aging mitochondria produce more reactive oxygen species (ROS) that can cause oxidative stress to nucleic acids, lipids and proteins. Therefore, mitochondrial autophagy is an indispensable degradation system of the body, which can maintain the survival of cells by selectively removing defective mitochondria that produce

excessive ROS [85]. The accumulation of a deal of misfolded proteins in the endoplasmic reticulum would damage the function of the endoplasmic reticulum and cause endoplasmic reticulum stress, so the endoplasmic reticulum autophagy acts a key role in the morphology and quality of the endoplasmic reticulum [86]. Therefore, we speculate whether autophagy caused by extracellular vesicles is a special autophagy differing from mitochondrial autophagy or endoplasmic reticulum autophagy and has a decisive effect on the content as well as the quantity of extracellular vesicles. Just saying this, the extracellular vesicles can be degraded by the autophagolysosomal pathway. However, in addition to the autophagy lysosomal pathway, intracellular degradation pathways also include the ubiquitin-proteolytic pathway, the caspase pathway etc. Therefore, the question of whether the extracellular vesicle can be degraded by these pathways requires further research.

The role of autophagy induced by extracellular vesicles

After the phenomenon that extracellular vesicles can be internalized and activate intracellular autophagy is discovered, we further discuss the function of autophagy caused by extracellular vesicles. Some studies have demonstrated that autophagy has the opposite effect in different pathological contexts.

Beneficial effect

In the past few decades, cardiovascular disease (CVD) has been a major concern in the world. Among the multiple CVD, acute myocardial infarction is the main cause of death [87]. Mesenchymal stem cell-derived exosomes activated autophagy in H9C2 cardiomyocytes and reduce cardiomyocyte apoptosis after H_2O_2 exposure [39]. Microvesicles with a diameter of 30 to 1000 nm were found to be involved in a variety of cancer processes [88]. The microvesicles secreted by human embryonic stem cell derived-mesenchymal stem cells significantly up-regulated the autophagy activity of K562 and HL60 cells and promoted apoptosis of leukemia cells, in other words, inhibited proliferation of tumor cells [89]. Other studies have found that ezetimibe can reduce liver steatosis and fibrosis by activating autophagy [49]. The above findings can prove that moderate autophagy caused by extracellular vesicles can play a protective role, which also provides a novel idea for the treatment of diseases.

Harmful effect

Under physiological conditions, autophagy undergoes orderly degradation of excess components in the cytosol by lysosomal means. In contrast, under certain pathological situations, inappropriate autophagy can damage normal cells and lead to aging and death [90]. Exosomes derived from acute myocardial infarction caused autophagy disorder in target cells by transmitting miR-30a. Nonetheless, inhibition of miR-30a in exosomes significantly reduced cardiomyocyte apoptosis [62]. Therefore, we speculate that the effect of extracellular vesicles on autophagy depends on its parental cells and the conditions that stimulate the production of extracellular vesicles.

Summarize and perspective

In this review, we summarize the ability of extracellular vesicles to active intracellular autophagy lysosome pathways after being internalized by various cells. This ability to active autophagy of extracellular vesicles is achieved by activating intracellular autophagy-related signaling pathways or by transferring active substances. After the extracellular vesicles activate autophagy of the target cells, the entire cytoplasm is enveloped in autophagosomes. Subsequently, the multivesicular body is delivered to the lysosome and the intraluminal vesicles such as exosomes are rapidly degraded by hydrolases. We also found that autophagy achieved by extracellular vesicles can dramatically reduces cell damage and abnormal cell proliferation under stress and tumor. We hypothesize that this protective effect is due to the degradation of extracellular vesicles containing harmful substances. The findings described in this review provide a novel view into the powerful therapeutic effect of EVs and EVs-induced autophagy. Although this curative effect has been discovered by numerous researchers, the molecular mechanisms involved in autophagy by active substances in extracellular vesicles are unclear. We must clearly understand that current technology does not distinguish exosomes from microparticles well. At the same time, whether the degradation of extracellular vesicles can directly determine the fate of cells

requires further exploration in the study of extracellular vesicles. Therefore, additional research is warranted to verify the potential issues. In any case, the therapeutic effects of autophagy caused by extracellular vesicles cannot be ignored and these findings may lead to a better disease treatment strategy to protect normal tissue cells from stress damage.

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Disclosure of conflict of interest

None.

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References

[1] Fischer D, Bussow J, Meybohm P, Weber CF, Zacharowski K, Urbschat A, Muller MM and Jennewein C. Microparticles from stored red blood cells enhance procoagulant and proinflammatory activity. Transfusion 2017; 57: 2701-2711.

[2] Kumar A, Stoica BA, Loane DJ, Yang M, Abulwerdi G, Khan N, Kumar A, Thom SR and Faden AI. Microglial-derived microparticles mediate neuroinflammation after traumatic brain injury. J Neuroinflammation 2017; 14: 47.

[3] Tual-Chalot S, Fatoumata K, Priou P, Trzepizur W, Gaceb A, Contreras C, Prieto D, Martinez MC, Gagnadoux F and Andriantsitohaina R. Circulating microparticles from patients with obstructive sleep apnea enhance vascular contraction: mandatory role of the endothelium. Am J Pathol 2012; 181: 1473-1482.

[4] Hayon Y, Dashevsky O, Shai E, Brill A, Varon D and Leker RR. Platelet microparticles induce angiogenesis and neurogenesis after cerebral ischemia. Curr Neurovasc Res 2012; 9: 185-192.

[5] Jia L, Fan J, Cui W, Liu S, Li N, Lau WB, Ma X, Du J, Nie S and Wei Y. Endothelial cell-derived microparticles from patients with obstructive sleep apnea hypoxia syndrome and coronary artery disease increase aortic endothelial cell dysfunction. Cell Physiol Biochem 2017; 43: 2562-2570.

[6] Li Z, Wang J and Yang X. Functions of autophagy in pathological cardiac hypertrophy. Int J Biol Sci 2015; 11: 672-678.

[7] Minakaki G, Menges S, Kittel A, Emmanouilidou E, Schaeffner I, Barkovits K, Bergmann A, Rockenstein E, Adame A, Marxreiter F, Mollenhauer B, Galasko D, Buzas EI, Schlotzer-Schrehardt U, Marcus K, Xiang W, Lie DC, Vekrellis K, Masliah E, Winkler J and Klucken J. Autophagy inhibition promotes SNCA/alpha-synuclein release and transfer via extracellular vesicles with a hybrid autophagosome-exosome-like phenotype. Autophagy 2018; 14: 98-119.

[8] Zhang N, Yang X, Yuan F, Zhu WG and Zhao Y. Autophagy-deficient tumor cells rely on extracellular amino acids to survive upon glutamine deprivation. Autophagy 2018; 14: 1652-1653.

[9] Gan-Or Z, Dion PA and Rouleau GA. Genetic perspective on the role of the autophagy-lysosome pathway in Parkinson disease. Autophagy 2015; 11: 1443-1457.

[10] Milton-Laskibar I, Aguirre L, Etxeberria U, Milagro FI, Martinez JA and Portillo MP. Involvement of autophagy in the beneficial effects of resveratrol in hepatic steatosis treatment. A comparison with energy restriction. Food Funct 2018; 9: 4207-4215.

[11] Giampieri F, Afrin S, Forbes-Hernandez TY, Gasparrini M, Cianciosi D, Reboredo-Rodriguez P, Varela-Lopez A, Quiles JL and Battino M. Autophagy in human health and disease: novel therapeutic opportunities. Antioxid Redox Signal 2019; 30: 577-634.

[12] Vegliante R and Ciriolo MR. Autophagy and autophagic cell death: uncovering new mechanisms whereby dehydroepiandrosterone promotes beneficial effects on human health. Vitam Horm 2018; 108: 273-307.

[13] Ren X, Chen Y, Peng H, Fang X, Zhang X, Chen Q, Wang X, Yang W and Sha X. Blocking autophagic flux enhances iron oxide nanoparticle photothermal therapeutic efficiency in cancer treatment. ACS Appl Mater Interfaces 2018; 10: 27701-27711.

[14] Song S, Tan J, Miao Y and Zhang Q. Effect of different levels of intermittent hypoxia on autophagy of hippocampal neurons. Sleep Breath 2017; 21: 791-798.

[15] Yu SY, Dong B, Fang ZF, Hu XQ, Tang L and Zhou SH. Knockdown of lncRNA AK139328 alleviates myocardial ischaemia/reperfusion injury in diabetic mice via modulating miR-204-3p and inhibiting autophagy. J Cell Mol Med 2018; 22: 4886-4898.

[16] Karnati HK, Garcia JH, Tweedie D, Becker RE, Kapogiannis D and Greig NH. Neuronal en-

riched extracellular vesicle proteins as biomarkers for brain traumatic injury. J Neurotrauma 2018; 36: 975-987.
[17] Faille D, El-Assaad F, Mitchell AJ, Alessi MC, Chimini G, Fusai T, Grau GE and Combes V. Endocytosis and intracellular processing of platelet microparticles by brain endothelial cells. J Cell Mol Med 2012; 16: 1731-1738.
[18] Rong H, Fan Y, Yang M, Zhang B, Sun D, Zhao Z, Wang D, Fan W, Wang J, Gu G, Li F, Liu X, Rao C, Chen H, Wang Y, Tian Y and Zhang J. Brain-derived microparticles activate microglia/macrophages and induce neuroinflammation. Brain Res 2018; 1694: 104-110.
[19] Tung SL, Boardman DA, Sen M, Letizia M, Peng Q, Cianci N, Dioni L, Carlin LM, Lechler R, Bollati V, Lombardi G and Smyth LA. Regulatory T cell-derived extracellular vesicles modify dendritic cell function. Sci Rep 2018; 8: 6065.
[20] Wang Y, Zhang L, Li Y, Chen L, Wang X, Guo W, Zhang X, Qin G, He SH, Zimmerman A, Liu Y, Kim IM, Weintraub NL and Tang Y. Exosomes/microvesicles from induced pluripotent stem cells deliver cardioprotective miRNAs and prevent cardiomyocyte apoptosis in the ischemic myocardium. Int J Cardiol 2015; 192: 61-69.
[21] Gu H, Liu Z, Li Y, Xie Y, Yao J, Zhu Y, Xu J, Dai Q, Zhong C, Zhu H, Ding S and Zhou L. Serum-derived extracellular vesicles protect against acute myocardial infarction by regulating miR-21/PDCD4 signaling pathway. Front Physiol 2018; 9: 348.
[22] Dasgupta SK, Le A, Chavakis T, Rumbaut RE and Thiagarajan P. Developmental endothelial locus-1 (Del-1) mediates clearance of platelet microparticles by the endothelium. Circulation 2012; 125: 1664-1672.
[23] Zhou Y, Cai W, Zhao Z, Hilton T, Wang M, Yeon J, Liu W, Zhang F, Shi FD, Wu X, Thiagarajan P, Li M, Zhang J and Dong JF. Lactadherin promotes microvesicle clearance to prevent coagulopathy and improves survival of severe TBI mice. Blood 2018; 131: 563-572.
[24] Jansen F, Yang X, Hoyer FF, Paul K, Heiermann N, Becher MU, Abu Hussein N, Kebschull M, Bedorf J, Franklin BS, Latz E, Nickenig G and Werner N. Endothelial microparticle uptake in target cells is annexin I/phosphatidylserine receptor dependent and prevents apoptosis. Arterioscler Thromb Vasc Biol 2012; 32: 1925-1935.
[25] Geng K, Kumar S, Kimani SG, Kholodovych V, Kasikara C, Mizuno K, Sandiford O, Rameshwar P, Kotenko SV and Birge RB. Requirement of gamma-carboxyglutamic acid modification and phosphatidylserine binding for the activation of Tyro3, Axl, and Mertk receptors by growth arrest-specific 6. Front Immunol 2017; 8: 1521.
[26] Mohning MP, Thomas SM, Barthel L, Mould KJ, McCubbrey AL, Frasch SC, Bratton DL, Henson PM and Janssen WJ. Phagocytosis of microparticles by alveolar macrophages during acute lung injury requires MerTK. Am J Physiol Lung Cell Mol Physiol 2018; 314: L69-L82.
[27] Shentu TP, Huang TS, Cernelc-Kohan M, Chan J, Wong SS, Espinoza CR, Tan C, Gramaglia I, van der Heyde H, Chien S and Hagood JS. Thy-1 dependent uptake of mesenchymal stem cell-derived extracellular vesicles blocks myofibroblastic differentiation. Sci Rep 2017; 7: 18052.
[28] Kim Y, Abplanalp WA, Jung AD, Schuster RM, Lentsch AB, Gulbins E, Caldwell CC and Pritts TA. Endocytosis of red blood cell microparticles by pulmonary endothelial cells is mediated by Rab5. Shock 2018; 49: 288-294.
[29] Costa Verdera H, Gitz-Francois JJ, Schiffelers RM and Vader P. Cellular uptake of extracellular vesicles is mediated by clathrin-independent endocytosis and macropinocytosis. J Control Release 2017; 266: 100-108.
[30] Guo S, Liang X, Guo M, Zhang X and Li Z. Migration inhibition of water stress proteins from Nostoc commune Vauch. via activation of autophagy in DLD-1 cells. Int J Biol Macromol 2018; 119: 669-676.
[31] Buckles TC, Ziemba BP, Masson GR, Williams RL and Falke JJ. Single-molecule study reveals how receptor and ras synergistically activate PI3Kalpha and PIP3 signaling. Biophys J 2017; 113: 2396-2405.
[32] Heras-Sandoval D, Perez-Rojas JM, Hernandez-Damian J and Pedraza-Chaverri J. The role of PI3K/AKT/mTOR pathway in the modulation of autophagy and the clearance of protein aggregates in neurodegeneration. Cell Signal 2014; 26: 2694-2701.
[33] Ebrahim N, Ahmed IA, Hussien NI, Dessouky AA, Farid AS, Elshazly AM, Mostafa O, Gazzar WBE, Sorour SM, Seleem Y, Hussein AM and Sabry D. Mesenchymal stem cell-derived exosomes ameliorated diabetic nephropathy by autophagy induction through the mTOR signaling pathway. Cells 2018; 7.
[34] Zhao GS, Gao ZR, Zhang Q, Tang XF, Lv YF, Zhang ZS, Zhang Y, Tan QL, Peng DB, Jiang DM and Guo QN. TSSC3 promotes autophagy via inactivating the Src-mediated PI3K/Akt/mTOR pathway to suppress tumorigenesis and metastasis in osteosarcoma, and predicts a favorable prognosis. J Exp Clin Cancer Res 2018; 37: 188.
[35] Pattingre S, Espert L, Biard-Piechaczyk M and Codogno P. Regulation of macroautophagy by mTOR and Beclin 1 complexes. Biochimie 2008; 90: 313-323.

[36] Xue R, Meng Q, Lu D, Liu X, Wang Y and Hao J. Mitofusin2 induces cell autophagy of pancreatic cancer through inhibiting the PI3K/Akt/mTOR signaling pathway. Oxid Med Cell Longev 2018; 2018: 2798070.

[37] Xin Y, Wan B, Yang Y, Cui XJ, Xie YC and Guo LH. Perfluoroalkyl acid exposure induces protective mitochondrial and endoplasmic reticulum autophagy in lung cells. Arch Toxicol 2018; 92: 3131-3147.

[38] Arslan F, Lai RC, Smeets MB, Akeroyd L, Choo A, Aguor EN, Timmers L, van Rijen HV, Doevendans PA, Pasterkamp G, Lim SK and de Kleijn DP. Mesenchymal stem cell-derived exosomes increase ATP levels, decrease oxidative stress and activate PI3K/Akt pathway to enhance myocardial viability and prevent adverse remodeling after myocardial ischemia/reperfusion injury. Stem Cell Res 2013; 10: 301-312.

[39] Liu L, Jin X, Hu CF, Li R, Zhou Z and Shen CX. Exosomes derived from mesenchymal stem cells rescue myocardial ischaemia/reperfusion injury by inducing cardiomyocyte autophagy Via AMPK and Akt pathways. Cell Physiol Biochem 2017; 43: 52-68.

[40] Shen S, Niso-Santano M, Adjemian S, Takehara T, Malik SA, Minoux H, Souquere S, Marino G, Lachkar S, Senovilla L, Galluzzi L, Kepp O, Pierron G, Maiuri MC, Hikita H, Kroemer R and Kroemer G. Cytoplasmic STAT3 represses autophagy by inhibiting PKR activity. Mol Cell 2012; 48: 667-680.

[41] Zhu Y, Wang Q, Tang X, Yao G and Sun L. Mesenchymal stem cells enhance autophagy of human intrahepatic biliary epithelial cells in vitro. Cell Biochem Funct 2018; 36: 280-287.

[42] Yang B, Xue Q, Qi X, Wang X, Jia P, Chen S, Wang T, Xue T and Wang J. Autophagy enhances the replication of Peste des petits ruminants virus and inhibits caspase-dependent apoptosis in vitro. Virulence 2018; 9: 1176-1194.

[43] Chiang WC, Wei Y, Kuo YC, Wei S, Zhou A, Zou Z, Yehl J, Ranaghan MJ, Skepner A, Bittker JA, Perez JR, Posner BA and Levine B. High-throughput screens to identify autophagy inducers that function by disrupting beclin 1/Bcl-2 binding. ACS Chem Biol 2018; 13: 2247-2260.

[44] Qu Y, Zhang Q, Cai X, Li F, Ma Z, Xu M and Lu L. Exosomes derived from miR-181-5p-modified adipose-derived mesenchymal stem cells prevent liver fibrosis via autophagy activation. J Cell Mol Med 2017; 21: 2491-2502.

[45] Andrade-Silva M, Cenedeze MA, Perandini LA, Felizardo RJF, Watanabe IKM, Agudelo JSH, Castoldi A, Gonçalves GM, Origassa CST, Semedo P, Hiyane MI, Foresto-Neto O, Malheiros DMAC, Reis MA, Fujihara CK, Zatz R, Pacheco-Silva A, Câmara NOS, de Almeida DC. TLR2 and TLR4 play opposite role in autophagy associated with cisplatin-induced acute kidney injury. Clin Sci (Lond) 2018; 132: 1725-1739.

[46] Wen X, Han XR, Wang YJ, Wang S, Shen M, Zhang ZF, Fan SH, Shan Q, Wang L, Li MQ, Hu B, Sun CH, Wu DM, Lu J and Zheng YL. MicroRNA-421 suppresses the apoptosis and autophagy of hippocampal neurons in epilepsy mice model by inhibition of the TLR/MYD88 pathway. J Cell Physiol 2018; 233: 7022-7034.

[47] Shin DM, Yuk JM, Lee HM, Lee SH, Son JW, Harding CV, Kim JM, Modlin RL and Jo EK. Mycobacterial lipoprotein activates autophagy via TLR2/1/CD14 and a functional vitamin D receptor signalling. Cell Microbiol 2010; 12: 1648-1665.

[48] Alvarez-Jimenez VD, Leyva-Paredes K, Garcia-Martinez M, Vazquez-Flores L, Garcia-Paredes VG, Campillo-Navarro M, Romo-Cruz I, Rosales-Garcia VH, Castaneda-Casimiro J, Gonzalez-Pozos S, Hernandez JM, Wong-Baeza C, Garcia-Perez BE, Ortiz-Navarrete V, Estrada-Parra S, Serafin-Lopez J, Wong-Baeza I, Chacon-Salinas R and Estrada-Garcia I. Extracellular vesicles released from mycobacterium tuberculosis-infected neutrophils promote macrophage autophagy and decrease intracellular mycobacterial survival. Front Immunol 2018; 9: 272.

[49] Kim SH, Kim G, Han DH, Lee M, Kim I, Kim B, Kim KH, Song YM, Yoo JE, Wang HJ, Bae SH, Lee YH, Lee BW, Kang ES, Cha BS and Lee MS. Ezetimibe ameliorates steatohepatitis via AMP activated protein kinase-TFEB-mediated activation of autophagy and NLRP3 inflammasome inhibition. Autophagy 2017; 13: 1767-1781.

[50] Xie H, Xu Q, Jia J, Ao G, Sun Y, Hu L, Alkayed NJ, Wang C and Cheng J. Hydrogen sulfide protects against myocardial ischemia and reperfusion injury by activating AMP-activated protein kinase to restore autophagic flux. Biochem Biophys Res Commun 2015; 458: 632-638.

[51] Zhao M, Sun L, Yu XJ, Miao Y, Liu JJ, Wang H, Ren J and Zang WJ. Acetylcholine mediates AMPK-dependent autophagic cytoprotection in H9c2 cells during hypoxia/reoxygenation injury. Cell Physiol Biochem 2013; 32: 601-613.

[52] Sooro MA, Zhang N and Zhang P. Targeting EGFR-mediated autophagy as a potential strategy for cancer therapy. Int J Cancer 2018; 143: 2116-2125.

[53] Song M, Wang Y, Shang ZF, Liu XD, Xie DF, Wang Q, Guan H and Zhou PK. Bystander autophagy mediated by radiation-induced exosomal miR-7-5p in non-targeted human bronchial epithelial cells. Sci Rep 2016; 6: 30165.

[54] Zhang Q, Shang M, Zhang M, Wang Y, Chen Y, Wu Y, Liu M, Song J and Liu Y. Microvesicles derived from hypoxia/reoxygenation-treated

human umbilical vein endothelial cells promote apoptosis and oxidative stress in H9c2 cardiomyocytes. BMC Cell Biol 2016; 17: 25.
[55] D'Souza-Schorey C and Schorey JS. Regulation and mechanisms of extracellular vesicle biogenesis and secretion. Essays Biochem 2018; 62: 125-133.
[56] Liu J, Jiang M, Deng S, Lu J, Huang H, Zhang Y, Gong P, Shen X, Ruan H, Jin M and Wang H. miR-93-5p-containing exosomes treatment attenuates acute myocardial infarction-induced myocardial damage. Mol Ther Nucleic Acids 2018; 11: 103-115.
[57] Chen J, Yu Y, Li S, Liu Y, Zhou S, Cao S, Yin J and Li G. MicroRNA-30a ameliorates hepatic fibrosis by inhibiting Beclin-1-mediated autophagy. J Cell Mol Med 2017; 21: 3679-3692.
[58] Saultz JN, Freud AG and Mundy-Bosse BL. MicroRNA regulation of natural killer cell development and function in leukemia. Mol Immunol 2018; [Epub ahead of print].
[59] Choi DC, Yoo M, Kabaria S and Junn E. MicroRNA-7 facilitates the degradation of alpha-synuclein and its aggregates by promoting autophagy. Neurosci Lett 2018; 678: 118-123.
[60] Jiang M, Wang H, Jin M, Yang X, Ji H, Jiang Y, Zhang H, Wu F, Wu G, Lai X, Cai L, Hu R, Xu L and Li L. Exosomes from MiR-30d-5p-ADSCs reverse acute ischemic stroke-induced, autophagy-mediated brain injury by promoting M2 microglial/macrophage polarization. Cell Physiol Biochem 2018; 47: 864-878.
[61] Li L, Wang Z, Hu X, Wan T, Wu H, Jiang W and Hu R. Human aortic smooth muscle cell-derived exosomal miR-221/222 inhibits autophagy via a PTEN/Akt signaling pathway in human umbilical vein endothelial cells. Biochem Biophys Res Commun 2016; 479: 343-350.
[62] Yang Y, Li Y, Chen X, Cheng X, Liao Y and Yu X. Exosomal transfer of miR-30a between cardiomyocytes regulates autophagy after hypoxia. J Mol Med (Berl) 2016; 94: 711-724.
[63] Cai S, Shi GS, Cheng HY, Zeng YN, Li G, Zhang M, Song M, Zhou PK, Tian Y, Cui FM and Chen Q. Exosomal miR-7 mediates bystander autophagy in lung after focal brain irradiation in mice. Int J Biol Sci 2017; 13: 1287-1296.
[64] Yan Q, Song Y, Zhang L, Chen Z, Yang C, Liu S, Yuan X, Gao H, Ding G and Wang H. Autophagy activation contributes to lipid accumulation in tubular epithelial cells during kidney fibrosis. Cell Death Discov 2018; 5: 2.
[65] Lapierre LR, Silvestrini MJ, Nunez L, Ames K, Wong S, Le TT, Hansen M and Melendez A. Autophagy genes are required for normal lipid levels in C. elegans. Autophagy 2013; 9: 278-286.
[66] Martin-Rincon M, Morales-Alamo D and Calbet JAL. Exercise-mediated modulation of autophagy in skeletal muscle. Scand J Med Sci Sports 2018; 28: 772-781.
[67] Fazeli G and Wehman AM. Safely removing cell debris with LC3-associated phagocytosis. Biol Cell 2017; 109: 355-363.
[68] Kulkarni R, Bajaj M, Ghode S, Jalnapurkar S, Limaye L and Kale VP. Intercellular transfer of microvesicles from young mesenchymal stromal cells rejuvenates aged murine hematopoietic stem cells. Stem Cells 2018; 36: 420-433.
[69] Pozuelo-Rubio M. 14-3-3 proteins are regulators of autophagy. Cells 2012; 1: 754-773.
[70] Jia H, Liu W, Zhang B, Wang J, Wu P, Tandra N, Liang Z, Ji C, Yin L, Hu X, Yan Y, Mao F, Zhang X, Yu J, Xu W and Qian H. HucMSC exosomes-delivered 14-3-3zeta enhanced autophagy via modulation of ATG16L in preventing cisplatin-induced acute kidney injury. Am J Transl Res 2018; 10: 101-113.
[71] Pozuelo-Rubio M. Regulation of autophagic activity by 14-3-3zeta proteins associated with class III phosphatidylinositol-3-kinase. Cell Death Differ 2011; 18: 479-492.
[72] DeYoung MP, Horak P, Sofer A, Sgroi D and Ellisen LW. Hypoxia regulates TSC1/2-mTOR signaling and tumor suppression through REDD1-mediated 14-3-3 shuttling. Genes Dev 2008; 22: 239-251.
[73] Weerasekara VK, Panek DJ, Broadbent DG, Mortenson JB, Mathis AD, Logan GN, Prince JT, Thomson DM, Thompson JW and Andersen JL. Metabolic-stress-induced rearrangement of the 14-3-3zeta interactome promotes autophagy via a ULK1- and AMPK-regulated 14-3-3zeta interaction with phosphorylated Atg9. Mol Cell Biol 2014; 34: 4379-4388.
[74] Wang J, Jia H, Zhang B, Yin L, Mao F, Yu J, Ji C, Xu X, Yan Y, Xu W and Qian H. HucMSC exosome-transported 14-3-3zeta prevents the injury of cisplatin to HK-2 cells by inducing autophagy in vitro. Cytotherapy 2018; 20: 29-44.
[75] Raimondo S, Saieva L, Corrado C, Fontana S, Flugy A, Rizzo A, De Leo G and Alessandro R. Chronic myeloid leukemia-derived exosomes promote tumor growth through an autocrine mechanism. Cell Commun Signal 2015; 13: 8.
[76] You L, Mao L, Wei J, Jin S, Yang C, Liu H, Zhu L and Qian W. The crosstalk between autophagic and endo-/exosomal pathways in antigen processing for MHC presentation in anticancer T cell immune responses. J Hematol Oncol 2017; 10: 165.
[77] Ktistakis NT and Tooze SA. Digesting the expanding mechanisms of autophagy. Trends Cell Biol 2016; 26: 624-635.
[78] Guay C and Regazzi R. Exosomes as new players in metabolic organ cross-talk. Diabetes Obes Metab 2017; 19 Suppl 1: 137-146.

[79] Xu J, Camfield R and Gorski SM. The interplay between exosomes and autophagy - partners in crime. J Cell Sci 2018; 131.
[80] Baixauli F, Lopez-Otin C and Mittelbrunn M. Exosomes and autophagy: coordinated mechanisms for the maintenance of cellular fitness. Front Immunol 2014; 5: 403.
[81] Abels ER and Breakefield XO. Introduction to extracellular vesicles: biogenesis, RNA cargo selection, content, release, and uptake. Cell Mol Neurobiol 2016; 36: 301-312.
[82] Fader CM and Colombo MI. Multivesicular bodies and autophagy in erythrocyte maturation. Autophagy 2006; 2: 122-125.
[83] Abdulrahman BA, Abdelaziz DH and Schatzl HM. Autophagy regulates exosomal release of prions in neuronal cells. J Biol Chem 2018; 293: 8956-8968.
[84] Fader CM, Sanchez D, Furlan M and Colombo MI. Induction of autophagy promotes fusion of multivesicular bodies with autophagic vacuoles in k562 cells. Traffic 2008; 9: 230-250.
[85] Civelek M, Mehrkens JF, Carstens NM, Fitzenberger E and Wenzel U. Inhibition of mitophagy decreases survival of caenorhabditis elegans by increasing protein aggregation. Mol Cell Biochem 2019; 452: 123-131.
[86] Wang Y, Zhao Z, Wei F, Luo Z and Duan Y. Combining autophagy-inducing peptides and brefeldin A delivered by perinuclear-localized mesoporous silica nanoparticles: a manipulation strategy for ER-phagy. Nanoscale 2018; 10: 8796-8805.
[87] Ding S, Fan Z, Lin C, Dai Q, Zhou J, Huang H, Xu Y and Zhong C. Therapeutic effects of ischemic-preconditioned exosomes in cardiovascular diseases. Adv Exp Med Biol 2017; 998: 271-281.
[88] Wu S, Ju GQ, Du T, Zhu YJ and Liu GH. Microvesicles derived from human umbilical cord Wharton's jelly mesenchymal stem cells attenuate bladder tumor cell growth in vitro and in vivo. PLoS One 2013; 8: e61366.
[89] Ji Y, Ma Y, Chen X, Ji X, Gao J, Zhang L, Ye K, Qiao F, Dai Y, Wang H, Wen X, Lin J and Hu J. Microvesicles released from human embryonic stem cell derived-mesenchymal stem cells inhibit proliferation of leukemia cells. Oncol Rep 2017; 38: 1013-1020.
[90] Zhu XM, Wang Q, Xing WW, Long MH, Fu WL, Xia WR, Jin C, Guo N, Xu DQ and Xu DG. PM2.5 induces autophagy-mediated cell death via NOS2 signaling in human bronchial epithelium cells. Int J Biol Sci 2018; 14: 557-564.