Canadian Journal of Cardiology ■ (2017) 1–10

Review

Autophagy, Metabolic Disease, and Pathogenesis of Heart Dysfunction

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ABSTRACT

In normal physiology, autophagy is recognized as a protective housekeeping mechanism that enables elimination of unhealthy organelles, protein aggregates, and invading pathogens, as well as recycling cell components and producing new building blocks and energy for cellular renovation and homeostasis. However, overactive or depressed autophagy is often associated with the pathogenesis of multiple disorders, including cardiac disease. During metabolic disorders, such as diabetes and obesity, dysregulation of autophagy frequently leads to cell death, cardiomyopathy, and cardiac dysfunction. In this article, we summarize the current understanding of autophagy—its classification, progression, and regulation; its roles in both physiological and pathophysiological conditions; and the balance between autophagy and apoptosis. We also explore how dysregulation of autophagy leads to cell death in models of metabolic disease and its contributing factors—including nutrient state, hyperglycemia, dyslipidemia, insulin inefficiency, and oxidative stress—and outline some recent efforts to restore normal autophagy in pathophysiological states. This information could provide potential targets for the prevention of, or intervention in, cardiac failure in metabolic disorders such as diabetes and obesity.

RÉSUMÉ

En physiologie normale, l'autophagie est reconnue comme un mécanisme protecteur de nettoyage qui favorise l'élimination des organites malsains, des agrégats de protéines et des pathogènes envahissants, ainsi que le recyclage des constituants cellulaires et la production de nouveaux éléments constitutifs et d'énergie pour le renouvellement et l'homéostasie des cellules. Toutefois, l'autophagie hyperactive ou faible est souvent associée à la pathogenèse de multiples troubles, y compris la maladie cardiaque. Lors de troubles du métabolisme, comme le diabète et l'obésité, la dysrégulation de l'autophagie mène fréquemment à la mort cellulaire, à la cardiomyopathie et à la dysfonction cardiaque. Dans cet article, nous résumons les connaissances actuelles sur l'autophagie — sa classification, sa progression et sa régulation; ses rôles dans les affections physiologiques et physiopathologiques; l'équilibre entre l'autophagie et l'apoptose. Nous explorons également de quelle façon la dysrégulation de l'autophagie mène à la mort cellulaire dans des modèles de maladies métaboliques et les facteurs qui y contribuent — y compris l'état nutritionnel, l'hyperglycémie, la dyslipidémie, l'inefficacité de l'insuline et le stress oxydatif — et exposons brièvement quelques récents efforts pour rétablir l'autophagie normale lors d'états physiopathologiques. Ces informations pourraient fournir des cibles potentielles en matière de prévention de l'insuffisance cardiaque ou d'intervention lors d'insuffisance cardiaque en présence de troubles du métabolisme comme le diabète ou l'obésité.

In physiological settings, autophagy is generally viewed as a protective mechanism that oversees the recycling of defective organelles and protein aggregates, ultimately contributing to the maintenance of cellular homeostasis, whereas abnormal upregulation or downregulation of autophagy is a distinctive feature of various pathologic states.[1,2] Both excessive and impaired autophagy could lead to cellular destruction through apoptosis, and the complex interactions between autophagy and apoptosis are a subject of increasing attention.[3-5] Delineating the nature of these interactions may further illuminate questions concerning the observed bioenergetic changes accompanying pathologic situations and potentially lead to the development of novel interventions. This review discusses the role of autophagy—from its regulation to its physiological influence—in the heart in metabolic disorders, particularly diabetes.

Autophagy

Autophagy is a cellular degradation process in which cytoplasmic constituents are recycled by lysosomal enzymes for reuse.[6] In contrast to the ubiquitin-proteasome degradation system, during which specific protein substrates are selectively ubiquitinated for breakdown in the proteasome, the general autophagic process nonselectively degrades protein

Received for publication November 3, 2016. Accepted January 4, 2017.

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See page 7 for disclosure information.

http://dx.doi.org/10.1016/j.cjca.2017.01.002

aggregates and defective organelles as part of a protective homeostatic mechanism to maintain cell survival.[7] However, during autophagy, specific organelles and proteins can also be selectively targeted, mostly through p62, for breakdown by the autophagic process.[8] Mitophagy, 1 example of organelle-specific autophagy, is particularly important for the cardiomyocyte. In this process, defective or damaged mitochondria are primed for selective autophagic recognition through PINK1-Parkin signalling or by other mitophagy receptors and are subsequently degraded by acidic lysosomal hydrolases.[9-12]

The three classifications of autophagy are (1) microautophagy, in which cytoplasmic contents are directly engulfed by lysosomes; (2) chaperone-mediated autophagy, in which chaperone proteins help protein substrates translocate across the lysosomal membrane; and (3) macroautophagy, in which cytoplasmic substrates are sequestered inside autophagosomes and degraded by lysosomal enzymes.[2,13] Of these, macroautophagy (referred to in this review as "autophagy") has been the most studied.

Processes of autophagy

Initially discovered by genetic screens performed in yeast, there are at present more than 30 known autophagy-related (*Atg*) genes that are confirmed in knockout mouse and mammalian cell models, in addition to genes related to this process.[1,14] These Atg genes initiate the nucleation step (Fig. 1B) of phagophore (isolation membrane) formation by activating the class III phosphoinositide 3-kinase (PI3K)/vacuolar protein sorting (Vps34) complex, which forms a multiprotein complex along with Beclin-1, Atg14, and Vps15 (Fig. 1A).[15] Subsequently, 2 ubiquitin-like conjugation systems control the process of phagophore elongation (Fig. 1, C-E).[2,16-18] The first pathway (Fig. 1D) involves conjugation of ubiquitin-like protein Atg12 to Atg5 by way of successive reactions involving E1-like enzyme Atg7 and E2-like enzyme Atg10. The resulting Atg12-Atg5 conjugate, along with Atg16L1, forms the Atg12-Atg5-Atg16L1 complex, which initiates phagophore elongation both independently and by promoting conjugation of microtubule-associated protein 1A/1B-light chain 3 (LC3-I; Atg8 in yeast) with phosphatidylethanolamine (PE), to form LC3-II. In the second pathway (Fig. 1E), LC3 undergoes proteolytic processing by Atg4 and conjugation to PE by E1-like Atg7 and E2-like Atg3 enzymes. The resulting amide bond is formed with the hydrophilic head of PE. After nucleation by Atg genes and elongation by the Atg12-Atg5 and LC3 pathways, a double-membrane vesicular compartment—the autophagosome—is formed. In this process, cytosolic components, including damaged organelles, pathogenic material, and protein aggregates, become sequestered within autophagosomes. Fusion of autophagosomes with lysosomes results in the formation of autolysosomes (Fig. 1F), inside of which cargo are degraded by acidic hydrolases, and their breakdown products are released into the cytosol for use in future anabolic reactions (Fig. 1G).

Regulation of autophagy

Regulation of autophagy is primarily accomplished through adenosine monophosphate–activated kinase (AMPK) and the mammalian target of rapamycin (mTOR) (Fig. 1B).[17-19] Under normal conditions, protein degradation by the proteasome is the major method used to supply amino acid building blocks for cellular functions.[20] During starvation, however, autophagy plays a more prominent role in amino acid generation.[13] Nutrient deprivation, with its associated reduction in amino acids and energy, in addition to reduced oxygen or growth factor signalling, increases autophagy. This occurs through inhibition of the serine/threonine protein kinase mTOR (Fig. 1H), which, as part of the mTOR complex 1 (mTORC1), is a nutrient sensor and negative regulator of autophagy.[7] Under nutrient-rich conditions, mTORC1 is activated and suppresses the unc-51–like autophagy activating kinase 1 (ULK1) complex, terminating autophagy. In contrast, the serine/threonine kinase AMPK (Fig. 1I) is a highly conserved cellular energy sensor and positive regulator of autophagy. Hence, AMPK deletion or mutation in mice reduces their response to inducers of autophagy.[21,22] AMPK is rapidly activated by a high AMP/adenosine triphosphate (ATP) ratio as a result of exercise, nutrition deprivation, or ischemia and maintains energy balance by decreasing pathways that use energy and increasing those that produce energy, in which autophagy is included.[23] In response to low energy levels, AMPK activates ULK1 by phosphorylation. Activated ULK1 itself phosphorylates Beclin-1, thereby activating the Vps34–Beclin-1 complex and inducing autophagy.

Physiological and pathophysiological roles of autophagy

Autophagy has a role in maintaining homeostasis and, ultimately, in contributing to cell survival. Under normal conditions, autophagy participates in the recycling of cytosolic components as well as pathogen elimination. It has been established that a basal level of autophagy is beneficial for survival and contributes to cellular homeostasis.[4] Insufficient autophagy leads to the accumulation of defective organelles and long-lived proteins, which can hinder cell survival and lead to apoptotic cell death, as evidenced in many Atg gene knockout mouse models.[1] Conversely, although increased autophagy can be beneficial for cell survival in the short term, its excessive or chronic induction is linked to cell death and pathologic states.[3] Past a certain threshold, autophagic destruction of organelles and cytosolic components can result in autophagy-induced apoptosis.[5] One important determinant of cell survival is the level of mitochondrial stress: mild stress promotes adaptive autophagy, moderate stress activates apoptosis as a result of cytochrome c release from the mitochondrial intermembrane space, and extreme stress initiates necrosis caused by ATP depletion.[4,24] It is thus not surprising that deviations from a basal level of autophagy are commonly observed in pathologic conditions, because metabolic alterations feature prominently in the pathology of a variety of diseases.[19] For instance, elevation of autophagy in breast cancer cells may be instrumental in promoting tumor survival. In contrast, diabetes, obesity, and neurodegenerative diseases such as Alzheimer's disease are linked to autophagy inhibition.[1] It is evident that elucidating the pathophysiological roles of autophagy is necessary to further our understanding of the bioenergetic changes accompanying various disease states and could potentially lead to the discovery of novel therapies.

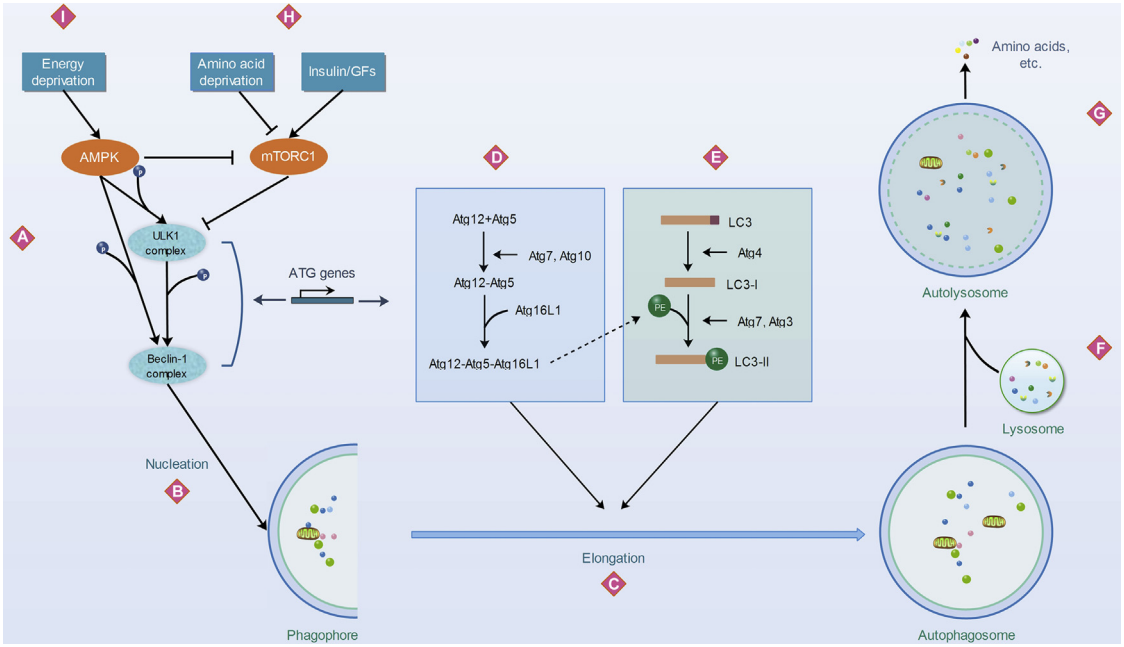

Figure 1. The macroautophagic process. (**A**) Mammalian target of rapamycin complex 1 (mTORC1), when activated by insulin and growth factors (GFs), inhibits the Unc-51–like autophagy activating kinase 1 (ULK1) complex to terminate autophagy. Amino acid deprivation directly inhibits mTORC1, releasing its block of the ULK1 complex, promoting autophagy. Adenosine monophosphate–activated kinase (AMPK), in response to energy deprivation and oxygen deprivation, is activated and inhibits mTORC1, in addition to activating the ULK1 complex directly through phosphorylation. (**B**) The activated ULK1 complex phosphorylates Beclin-1, which, along with autophagy-related (Atg)14/vacuolar protein sorting (Vps) 15/class III phosphoinositide 3-kinase (PI3K)/Vps34, leads to nucleation. In addition, AMPK can directly activate Beclin-1 through phosphorylation. (**C**-**E**) Elongation of the phagophore (isolation membrane) is accomplished through 2 systems. (**D**) In the first pathway, Atg12-Atg5 associates with Atg16L1 to form the Atg12-Atg5-Atg16L1 complex, promoting the conjugation of phosphatidylethanolamine (PE) with light chain 3 (LC3)-I. (**E**) In the second pathway, LC3 is proteolytically processed to LC3-I and then to LC3-II after conjugation to PE. p62 links LC3-II and polyubiquitinated proteins inside the autophagosome. (**F**) Fusion of autophagosomes with lysosomes to form autolysosomes results in degradation of its contents, and (**G**) the amino acids and other building blocks generated are released into the cytoplasm for reuse by the cell. (**H**, **I**) The 2 master regulators of autophagy, AMPK and mTORC1, are subject to control by energy deprivation, amino acid deprivation, and insulin/growth factor signalling.

Balance between autophagy and apoptosis

Inhibition of autophagy by both genetic manipulation and pharmacologic inhibition increases apoptosis, likely because of an intracellular accumulation of protein aggregates and damaged cytoplasmic components.[25] For instance, in lysosome-associated membrane protein 2 (LAMP2)-deficient cells, disrupted autophagosome-lysosome fusion leads to autophagosome accumulation and, consequently, cell death.[26] In addition to exhibiting disrupted autophagy in multiple organs, knockout of LAMP2 in mice also causes Danon disease, which counts fatal cardiomyopathy as 1 of its features.[27] In mice deficient in Atg5 or Atg7, mortality is observed within 1 day of delivery because of a disruption in the expected increase in autophagy, which is normally observed immediately after birth.[28,29] Additionally, mutant mice with neuron-specific depletion of Atg5 or Atg7 accumulate cytoplasmic protein aggregates and exhibit neurodegeneration.[30,31] Furthermore, a deficiency of Atg5 in T cells increases the apoptosis of mature T cells in peripheral organs.[32] Finally, homozygous Beclin-1$^{-/-}$ embryonic stem cells also show a severely altered autophagic response, with Beclin-1$^{-/-}$ embryos experiencing mortality in early embryogenesis.[33] In contrast, when autophagy is appropriately induced, cell death can be reduced. For example, autophagy promoted by glyceraldehyde-3-phosphate dehydrogenase can preserve cell survival even after apoptotic cytochrome c release.[34] Inhibition of mTOR, the "master negative regulator" of autophagy, can promote autophagy and consequently enable cell survival in several models of Huntington disease.[35] It should be noted that excessive autophagic destruction of cytoplasmic components and organelles leads to cell death. Hence, autophagic vacuoles are observed concurrently with apoptotic and necroptotic vacuoles in multiple models.[5]

One centralized mechanism that regulates the balance between autophagy and apoptosis is the interaction between antiapoptotic Bcl-2 proteins (in most cases Bcl-2 and Bcl-X_L) and Beclin-1, a key component of the autophagy-promoting Beclin-1–Vps34-Vps15 complex that is subject to regulation by a variety of proteins (Fig. 2).[36] On autophagy induction, Beclin-1 binds to its partners and initiates autophagy (Fig. 2A). However, binding of antiapoptotic Bcl-2 proteins to Beclin-1 disrupts this interaction, preventing autophagy (Fig. 2B). At the same time, this association of antiapoptotic Bcl-2 proteins and Beclin-1 also abrogates the usual antiapoptotic role of these Bcl-2 proteins. Thus, the relative concentration of antiapoptotic Bcl-2 proteins and Beclin-1, in addition to their mutual interaction, constitutes a rheostat for autophagy and cell death.[37] In this regard, phosphorylation of Beclin-1 by Mst1 (on Ser-90),[38] endothelial growth factor receptor (EGFR) (on Tyr-229, Tyr-233, and Tyr-352),[39] and the class

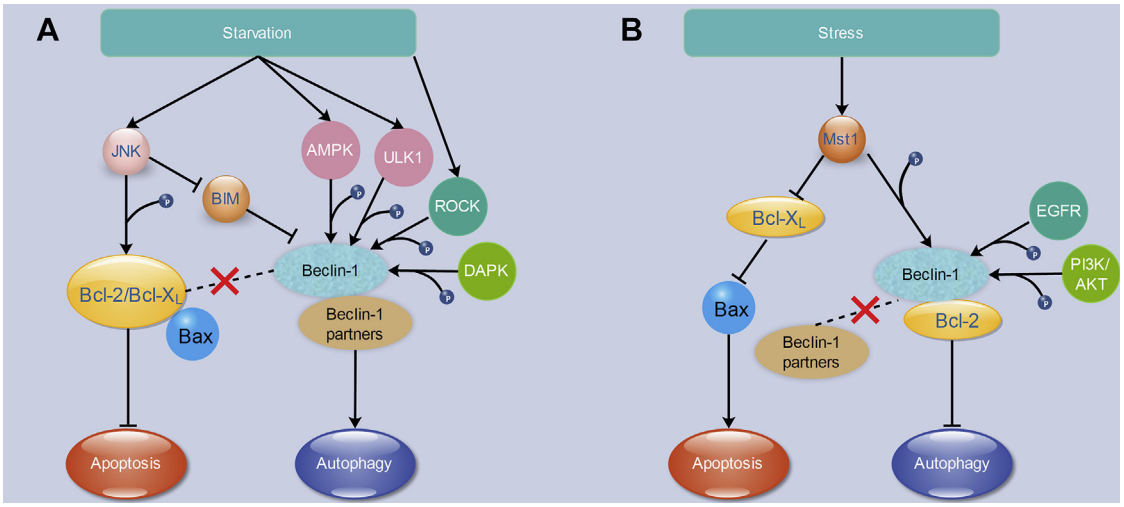

Figure 2. The balance between autophagy and apoptosis. (**A**) The association between Beclin-1 and Bcl-2 can be disrupted by either phosphorylation at a specific locus or by the competitive binding of other BH3-only proteins with Bcl-2/Bcl-X_L. JNK can be phosphorylated and activated by different stimuli, especially starvation. On activation, JNK can either phosphorylate Bcl-2 at multiple sites or, alternatively, phosphorylate Bcl-2–interacting mediator of cell death (BIM) to thereby dissociate its inhibitory interaction with Beclin-1; both situations cause dissociation between Bcl-2 and Beclin-1 and lead to induction of autophagy. Beclin-1 can also be phosphorylated by adenosine monophosphate–activated kinase/Unc-51–like autophagy activating kinase 1 (AMPK/ULK-1), rho associated coiled-coil containing protein kinase 1 (ROCK1), and death-associated protein kinase (DAPK), all of which result in dissociation of Beclin-1 from Bcl-2 and increased autophagy. Additionally, the interaction between Beclin-1 and Bcl-2 can be disrupted by a replacement interaction between Bcl-2 and other BH3-only proteins. However, it should be noted that some of these interactions may also disrupt the binding of Bcl-2 to Bax, leading to apoptosis. (**B**) Phosphorylation of Beclin-1 at specific sites can promote the association of Beclin-1 and Bcl-2. Phosphorylation and activation of Mst1 can inhibit autophagy by phosphorylating Beclin-1 and promoting its interaction with Bcl-2. Concurrently, Mst1 phosphorylation can abrogate the inhibitory effect of Bcl-X_L on apoptosis by phosphorylating Bcl-X_L at Ser-14 and dissociating Bcl-X_L from Bax. Beclin-1 is also subject to regulation by phosphoinositide 3-kinase (PI3K)/Akt and endothelial growth factor receptor (EGFR), in both cases leading to inhibition of autophagy.

I PI3K-Akt axis (on Ser-295) facilitates the interaction between Beclin-1 and antiapoptotic Bcl-2 proteins (in the case of the PI3K-Akt axis, the interaction is with 14-3-3 and vimentin intermediate filament proteins),[40] thereby inhibiting autophagy (Fig. 2B). In contrast, phosphorylation of antiapoptotic Bcl-2 proteins by JNK,[41] and phosphorylation of Beclin-1 by AMPK,[42] ULK1 (both on Ser-14),[43] ROCK1 (on Thr-119),[44] and death-associated protein kinase (DAPK) (on Thr-119),[45] dissociates the binding between Bcl-2 and Beclin-1, promoting autophagy (Fig. 2A). The interaction between Beclin-1 and antiapoptotic Bcl-2 proteins is also regulated by competitive binding between Beclin-1, other BH3-only proteins, antiapoptotic Bcl-2 proteins, and other Bcl-2 family members.[46-48] For example, Bcl-2–interacting mediator of cell death (BIM) interacts with Beclin-1 and facilitates its interaction with dynein light chain 1, thereby preventing autophagy (Fig. 2A).[48] JNK, which phosphorylates Bcl-2 and dissociates the interaction between Beclin-1 and Bcl-2, can also phosphorylate BIM and disrupt the BIM–Beclin-1 interaction, promoting autophagy induction (Fig. 2A).[48] It should be noted that other BH3-only proteins can also disrupt the interaction between Beclin-1 and Bcl-2 family proteins.[49-51]

Autophagy in the heart

A series of studies have shown that autophagy is related to cardiac disease, including dilated cardiomyopathy, valvular disease, and ischemic cardiac disease.[52] In fact, autophagic cell death has been detected more often than apoptotic cell death in multiple cardiac disease models.[53] For example, LAMP2-deficient mice showed abnormal cardiomyocyte ultrastructure and reduced contractility—characteristics related to Danon disease.[54] Loss of autophagy, through depletion of macrophage migration inhibitory factor (MIF) or myeloid cell leukemia 1 (MCL-1),[55,56] causes serious cardiomyopathy. Mortality in cardiac-specific Atg5-deficient mice, which display impaired sarcomere structure and dysfunctional mitochondria, is observed after 6 months.[57] Furthermore, temporal cardiac-specific ablation of Atg5 expression causes cardiac hypertrophy, left ventricular dilatation, and contractile dysfunction and also results in disorganized sarcomere structure and mitochondrial misalignment and aggregation.[58]

In contrast, GFP-LC3 transgenic mice showed increased numbers of autophagosomes (mitophagy), which is essential for mitochondria turnover and cardiomyocyte function.[59] In addition, multiple gene manipulation investigations have demonstrated that increased autophagy is associated with decreased apoptosis and ameliorated cardiac dysfunction in different cardiac disease models.[60-64]

Obesity, diabetes, and cardiomyocyte autophagy

In metabolic disorders such as obesity and diabetes,[65] there is prominent evidence of cardiac dysfunction, which has been linked to hyperglycemia, dyslipidemia, insulin deficiency, and oxidative stress, with defective autophagy being widely implicated in the pathogenesis of this cardiac dysfunction. Reduced levels of autophagy have been reported in obesity, likely as a consequence of elevated lipid, amino acid, and insulin levels in the circulation.[19,66,67] Mechanistically,

obesity can suppress autophagy by increasing mTORC1 activity, a process that is further aggravated during ischemia (Fig. 3A).[68-70] Rescue of cardiac autophagy by depletion of Akt2 or caspase recruitment domain family member 9 (CARD9) in animals with a high-fat diet (HFD)-induced obesity resulted in cardioprotective effects.[71,72] Interestingly, impaired autophagy has also been linked to cardiac insulin resistance, a phenomenon observed in both obesity and diabetes.[68] Reduced cardiac autophagy is also thought to be a contributing factor in diabetic cardiomyopathy, because restoration of autophagy by metformin ameliorates cardiac dysfunction in diabetic mice.[21] Hyperglycemia contributes to supressed autophagy, leading to protein aggregation and the accumulation of dysfunctional organelles (especially mitochondria), ultimately resulting in cardiomyocyte cell death.[73] Overall, it is accepted that the proper restoration of autophagy in cardiomyocytes would be beneficial in preventing the cardiomyopathy observed in obesity and diabetes. Hence, to overcome the associated cardiac insults that occur during these conditions, a better understanding of how autophagy is altered in these 2 pathologic states is necessary.

Nutrient state and autophagy

Nutrient starvation is the most widely investigated factor that contributes to autophagy. The 2 master regulators of autophagy, AMPK and mTOR, are both subject to regulation by nutrient deprivation (Fig. 3A). AMPK, a positive regulator of autophagy, is an energy sensor that can quickly respond to low-energy states by initiating autophagic machinery through inhibiting mTOR and activating ULK1 and Beclin-1.[17,23,74] Upstream signaling of AMPK may be involved in inducing autophagy in response to nutrient deprivation, with calcium signalling being an important contributor in this process.[75-77] However, whether or not this process is relevant in the heart needs further clarification. In contrast to AMPK, mTOR is a negative regulator of autophagy that is activated under nutrient-rich conditions, such as in chronic metabolic disorders, including obesity and diabetes (Fig. 3A).[2,14] Inhibition of mTOR with rapamycin improves cardiac function in mice with type 2 diabetes.[78] One typical example of nutrient supply disruption is the birth of neonates, during which autophagy is dramatically increased before the establishment of suckling.[28] Prevention of mTOR inhibition by Rag guanosine triphosphatase (GTPase) leads to defective autophagy and neonatal death, a phenotype similar to that observed in Atg5 or Atg7 knockout mice.[79] The association between mTOR and the downstream ULK1 complex is also nutrient dependent.[80]

Hyperglycemia and autophagy

One of the most consistently observed features in diabetes is hyperglycemia, which can dramatically alter the signalling pathways and metabolic patterns of the heart. Hyperglycemia suppresses autophagic flux in cardiomyocytes and enhances protein aggregation in both *in vivo* and *in vitro* models (Fig. 3A).[81] AMPK activation by metformin can greatly boost autophagy and improve cardiac structure and function; however, there is evidence that both high glucose *in vitro* and streptozotocin-induced hyperglycemia *in vivo* can inhibit AMPK activity in the cardiomyocyte.[21,42,73,82] Another protein implicated in hyperglycemia-suppressed autophagy is Mst1, which on activation promotes apoptosis and inhibits autophagy by promoting the association between Bcl-2 and Beclin-1.[38,83] Cardiomyocyte Mst1 can be phosphorylated and activated by both high glucose and diabetes. Furthermore, Mst1 overexpression increased, whereas Mst1 knockout decreased, cardiomyocyte apoptosis both *in vivo* and *in vitro*.[81] Restoration of autophagy can also attenuate high-glucose–induced cardiomyocyte cell death and cardiomyopathy by promoting the expression of Sirt1.[84] However, diabetes does not always suppress cardiac autophagy,[85,86] because this process also depends on the type and duration of diabetes and may involve factors other than hyperglycemia, including dyslipidemia, altered insulin signalling, oxygen deprivation, and oxidative stress.

Dyslipidemia and autophagy

Hallmarks of dyslipidemia include decreased high-density lipoprotein levels, increased triglyceride levels, and postprandial lipemia, all of which are frequently observed in diabetes and obesity and are important risk factors for the subsequent development of cardiac dysfunction. HFDs have been shown to disrupt autophagosome-lysosome fusion, leading to intracellular Ca^{2+} dysregulation, mitochondrial injury, apoptosis, cardiac hypertrophy, and cardiomyocyte contractile dysfunction (Fig. 3A).[71,87] Knockout of protein-tyrosine phosphatase 1B (PTP1B) can abrogate the detrimental effect of HFD in the mouse heart by restoring cardiomyocyte autophagy.[88] However, AMPK inhibition by compound C nullifies PTP1B knockout-conferred protection against HFD-induced cardiomyocyte anomalies, suggesting that the function of AMPK is dampened in HFD-suppressed autophagy. In another study, HFD mice also displayed decreased cardiac autophagy through deregulated cardiac activation of Rheb and mTORC1.[69] The resultant increase in injury after ischemia can be reversed by pharmacologic or genetic inhibition of mTORC1, implicating a role for mTOR in this process. Interestingly, a study in hypercholesterolemic swine demonstrated activated upstream and downstream mTOR signalling, inhibited autophagy, and increased cardiac hypertrophy markers, which together confirm the role of mTOR in high-fat, high-cholesterol diet–induced cardiac autophagy dysregulation.[89] Akt activation was also involved in this HFD-induced suppression of autophagy and the resulting cardiac anomalies.[87] Abrogating autophagy suppression by modulating these signalling pathways can alleviate HFD-induced cardiac dysfunction and may be a potential target in metabolic syndrome–induced heart disease.[69,71,87,88,90,91]

Growth factor/insulin signalling and autophagy

Deficiency of insulin action is another feature associated with diabetes and obesity. It is widely accepted that growth factor or insulin, or both, can activate mTOR and subsequently inhibit autophagy through Akt signalling (Fig. 3B).[92,93] Cardiac-specific deletion of the insulin receptor substrates Irs1 and Irs2 in cardiomyocytes led to reduced mTOR signalling, persistent overactivation of autophagy, increased apoptosis, and precipitated mitochondrial dysfunction and the subsequent development of cardiac failure.[94]

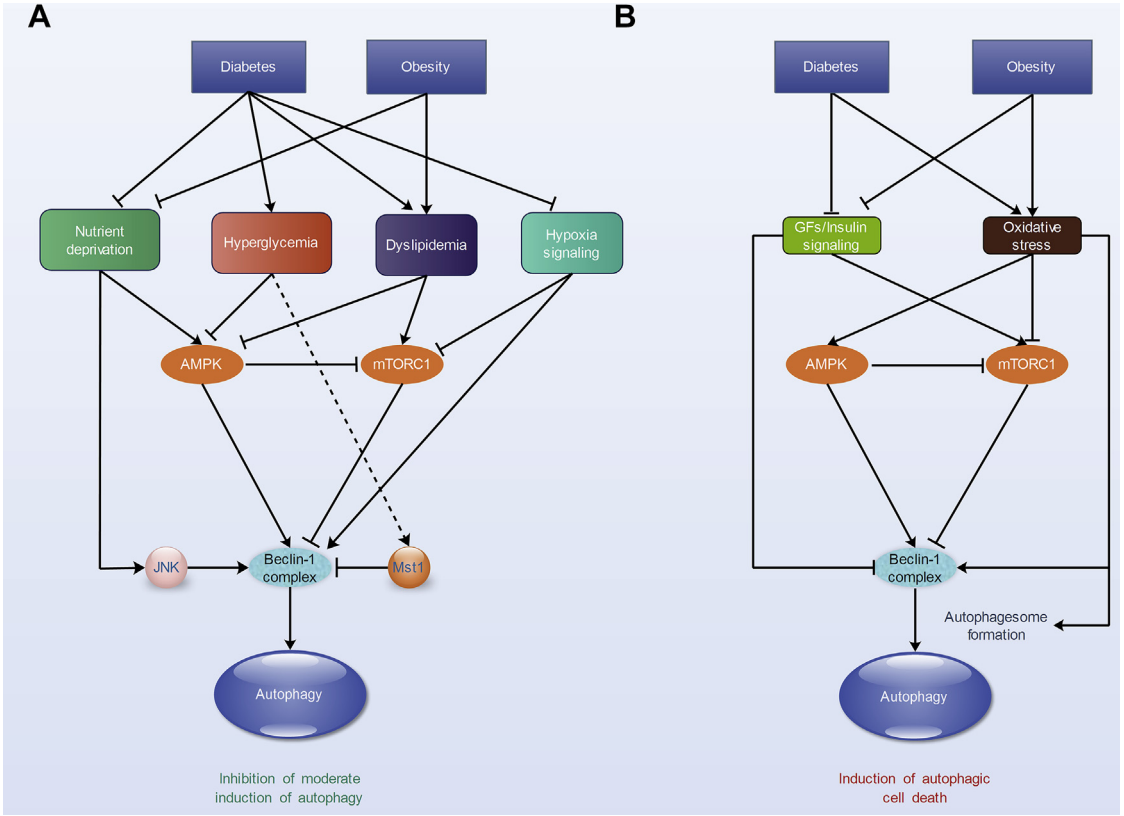

Figure 3. The regulation of autophagy in the heart in diabetes. (**A**) In diabetes and obesity, hyperglycemia, dyslipidemia, inhibition of starvation pathway or hypoxia signalling reduces induction of autophagy by modulating the activity of adenosine monophosphate–activated kinase (AMPK), mammalian target of rapamycin (mTOR), and Beclin-1. Hyperglycemia and nutrient status can also regulate the interaction between Beclin-1 and Bcl-2, therefore regulating autophagy by JNK and Mst1, respectively. (**B**) In the case of severe metabolic syndrome or other stresses such as ischemia/reperfusion, the inhibition of insulin signalling on autophagy is abrogated, and cell survival mechanisms are disrupted. Oxidative stress activates the AMPK-mTOR–Beclin-1 machinery and induces moderate to severe autophagy, leading to cell death.

This can be ameliorated by mTOR activation or genetic suppression of autophagy. However, the autophagy activation resulting from insulin resistance in diabetes models is often overshadowed by the effect of hyperglycemia in inhibiting autophagy. Consequently, more cases of autophagy suppression than autophagy induction are observed in models of diabetes. Therefore, the effects of insulin signalling on cardiomyocyte autophagy in the context of metabolic disorders require further investigation. Notwithstanding this uncertainty, it should be noted that exercise has been widely recognized as an inducer of autophagy in multiple organs.[95-97] An autophagy-defective mouse model, in which Atg7 is deleted in cardiac and skeletal muscle, displays a reduction in the beneficial effect of exercise in opposing insulin resistance in the heart. Conversely, overexpression of Atg7 and upregulation of autophagy ameliorates desmin-related cardiomyopathy.[98] A ULK1 knockout mouse model made by the same group also demonstrated that autophagy is essential for exercise-conferred cardiac protection against obesity and diabetes.[99] Besides Atg7 and ULK1, the interaction between Bcl-2 and Beclin-1 is essential for the induction of autophagy by exercise, because preventing disruption of the Bcl-2–Beclin-1 interaction by knockin of a Bcl-2 mutation abrogates exercise-induced autophagy in the heart.[95,96] AMPK, a well-established molecule that can be activated by exercise, is also essential in exercise-induced autophagy and participates in cardiac protection against ischemic injury.[21,22,97]

Oxygen deprivation, oxidative stress, and autophagy

The heart is an organ that is particularly sensitive to oxygen deficiency and oxidative stress.[100] Oxygen deprivation is a key factor in the pathogenesis of various cardiac diseases, including cardiac failure and myocardial infarction.[100] Hypoxia generally induces autophagy in cardiomyocytes (Fig. 3A)—a response that can be protective in some situations but detrimental when out of control. One of the mechanisms by which hypoxia can promote autophagy is through inducing BNIP3 and BNIP3L through hypoxia-inducible factor, which dissociates the interaction between Beclin-1 and Bcl-2/Bcl-X_L.[49] The induction of autophagy attenuates hypoxia-induced cardiomyocyte apoptosis and improves cardiac function through a variety of mechanisms, particularly by AMPK activation.[64,101-104] One possible caveat, however, is the use of an ischemia/reperfusion model in some of the preceding studies, for which the importance of oxidative stress is intensified. Nevertheless, autophagy activation has been observed in both ischemia/reperfusion and hypoxia alone (Fig. 3B) and can be either adaptive or maladaptive.[105] During diabetes or obesity, oxidative stress can be generated

by glucolipotoxicity or cardiac ischemia.[106] Although autophagy induction by gene overexpression has been shown to be protective against oxidative stress in the heart,[91,107] prolonged autophagy activation as a consequence of oxidative stress usually results in cardiomyocyte apoptosis and attendant cardiac disease.[108,109]

Concluding Remarks and Future Perspectives

Autophagy is a housekeeping process that links energy status and stress with cell survival mechanisms. The heart can quickly respond to stress—induced by abnormal intracellular accumulation of organelles or protein aggregates—or starvation by activating the autophagy machinery to respond to such insults and rescue the cell from death. However, the regulation of autophagy is often challenged during pathophysiological states, especially by chronic metabolic insults such as diabetes and obesity. In these situations, regulation of nutrient state, glucose levels, lipid levels, insulin signalling, oxygen availability, and oxidative stress are often disrupted, and the intricate balance between autophagy and apoptosis is lost. Numerous studies using gene manipulation (LAMP2, Atg5/7, Beclin-1, PTP1B, MIF, MCL-1, Akt2, CARD9, Mst1, LC3, and Rag GTPase), pharmacologic regulation (metformin, compound C, and rapamycin), and even lifestyle (diet and exercise) alterations have been conducted to explore the involvement of autophagy in the pathogenesis of cardiac dysfunction during diabetes and obesity. It has been suggested that the proper induction of autophagy may reduce cardiomyocyte cell death and cardiomyopathy, because a number of studies have demonstrated ameliorated cardiac dysfunction after promotion of autophagy. Collectively, these data provide potential targets for the prevention of, and intervention during, cardiac complications in patients with metabolic disorders. However, a full understanding of autophagy regulation and the cross talk between autophagy and apoptosis in the heart remains elusive. Whether autophagy is impaired or activated in certain disease models is still controversial, and the extent to which the effects of some interventions rely on autophagy pathways remains undetermined. Using some of our existing drugs (such as metformin and statin drugs) and promoting healthy lifestyle alterations (such as caloric restriction and exercise) as part of targeted interventions to restore autophagy and improve cardiac function in metabolic diseases is a potential direction of future research. Additionally, another particularly important organelle of interest in the heart is the mitochondrion, which provides sustainable energy for the beating heart and whose disruption in autophagy (mitophagy) has been emphasized in recent studies. In addition to exploring these issues, it may be helpful to also investigate mitophagy and its disruption in the heart in metabolic disorders to provide further insight into the cardiac health of obese and diabetic patients. Overall, the ramifications of restoring proper autophagy processes in the heart after bioenergetic changes related to metabolic disorders appear to be positive and constitute a promising direction for future study. The elucidation of these issues will serve to further our understanding of the pathophysiology of heart dysfunction and may potentially lead to the development of novel interventions toward the prevention or treatment, or both, of metabolic diseases.

Funding Sources

This work was supported by an operating grant from the Canadian Institutes of Health Research (CIHR-MOP-133547).

Disclosures

The authors have no conflicts of interest to disclose.

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