Targeting Autophagy for the Therapeutic Application of Histone Deacetylase Inhibitors in Ischemia/Reperfusion Heart Injury

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Ischemic heart disease is a leading cause of morbidity and mortality in the United States and other parts of the world. Despite therapeutic breakthroughs over the past decades such as percutaneous coronary intervention, antiplatelet and anti-thrombotic therapies, and angioplasty, the prevalence of ischemic heart diseases remains extremely high and constitutes a devastating factor for heart failure. Among various therapeutic strategies for ischemic heart disease, enormous efforts have been made to limit ischemia/reperfusion (I/R) injury, which occurs when the ischemic myocardium is reperfused with oxygen and substrate-rich blood, which paradoxically worsens heart function. Ischemic myocardium, with nutrient and oxygen deprivation and buildup of reactive oxygen species (ROS), uses glycolysis as the primary source of metabolic energy. As a consequence, metabolic acidosis, hyperkalemia, and Ca²⁺ overload develop in cardiomyocytes after coronary artery occlusion, leading not only to cardiomyocyte apoptosis during the acute phase but also to delayed adverse myocardial remodeling, which further compromises cardiac function. Therefore, limiting I/R-induced myocardial ROS accumulation and apoptosis benefits both short- and long-term survival and quality of life. Although the mechanism responsible for I/R-induced cardiac abnormalities has been focused largely on necrosis and type I (apoptotic) programmed cell death, an intriguing and provocative paradigm has emerged recently that highlights a unique role for dysregulated macroautophagy (hereafter referred to as autophagy) in the heart that may render cardiomyocytes more prone to I/R injury and long-term postinfarction cardiac remodeling, which further compromises cardiac function. It has been perceived that autophagy induced by ischemic preconditioning is essential for cardioprotection. To this end, new and innovative strategies to maintain or restore myocardial autophagy homeostasis and its attendant, cardiomyocyte survival, have been the subject of intensive investigation.

The Janus-Faced Role of Autophagy Induction in I/R Injury

Autophagy is a tightly regulated, lysosome-dependent catabolic process responsible for turnover of long-lived proteins and intracellular structures that are damaged or malfunctions. The evolutionally conserved bulk degradation process is turned on when cells experience stress, including nutrient and energy deprivation. The autophagic pathway consists of 4 distinct but consecutive steps: initiation, formation of autophagosomes (ie, the double-membrane structures that encircle cargo of damaged cytosolic constituents), generation of autophagolysosomes via docking and fusion with lysosomes, and final degradation of sequestered cargo. Sequestration of cytoplasmic cargo such as long-lived proteins, damaged organelles, and protein aggregates into the double-membrane vesicle autophagosomes occurs before fusion with lysosomes for degradation of its contents by acidic hydrolases. Although physiological levels of autophagy are essential for mitochondrial function, cell survival, and cell function, excessive activation of autophagy can be detrimental, leading ultimately to autophagic cell death. Recent findings identified an important role for autophagy in the pathogenesis of human diseases, in particular heart diseases, implicating the therapeutic potential of autophagy regulation against heart anomalies. In light of the indispensable role of autophagy for cardiac homeostasis, recent attention has focused on understanding the role of autophagy regulation in I/R injury. It has been demonstrated that autophagy seems to play a paradoxical role in I/R injury. In ischemia, induction of autophagy via AMP kinase is protective, whereas reperfusion stimulates autophagy with Beclin-1 upregulation to compromise cardiac cell survival and function. This dual regulatory paradigm of autophagy in the ischemia and reperfusion phases may underscore the homeostatic and drug intervention machinery for I/R heart injury. Further evidence indicates that I/R injury impairs autophagosome clearance (autophagy flux) mediated in part through an ROS-induced decline in lysosome-associated membrane protein-2 and upregulation of the autophagy initiation.

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protein Beclin-1, leading to the ultimate cardiomyocyte death. Recently, a number of pharmaceutical therapies targeting I/R injury have been designed to orchestrate multiple protein complexes and signaling pathways in autophagy. For instance, sevoflurane has been shown to offer cardioprotection through ROS-mediated upregulation of autophagy in I/R. On the contrary, in vitro evidence suggested that α-lipoic acid protects H9C2 myoblasts against hypoxia/reoxygenation injury through suppression of autophagy. The precise role of autophagy regulation contributing to cell survival and death in ischemic hearts remains controversial. In this issue of Circulation, Xie and colleagues report that suberoylanilide hydroxamic acid (SAHA; vorinostat), a histone deacetylase (HDAC) inhibitor approved by the Food and Drug Administration for cancer treatment, attenuated myocardial reperfusion injury in rabbits. These results revealed that SAHA reduced infarct size and partially rescued systolic function when administered either before surgery or at the time of reperfusion. SAHA was found to facilitate autophagic flux in the infarct border zone in rabbit myocardium and in mice harboring an red fluorescent protein-green fluorescent protein-LC3 transgene. In cultured myocytes subjected to I/R, SAHA overtly alleviated cell death, the effect of which was correlated with increased autophagy. The permissive role of autophagy in SAHA-related beneficial effects was consolidated by the mitigation of SAHA efficacy through RNAi knockdown of autophagy protein complexes and signaling pathways in autophagy. The permissive role of autophagy regulation contributing to cell survival and death in ischemic hearts remains controversial. In this issue of Circulation, Xie and colleagues report that suberoylanilide hydroxamic acid (SAHA; vorinostat), a histone deacetylase (HDAC) inhibitor approved by the Food and Drug Administration for cancer treatment, attenuated myocardial reperfusion injury in rabbits. These results revealed that SAHA reduced infarct size and partially rescued systolic function when administered either before surgery or at the time of reperfusion. SAHA was found to facilitate autophagic flux in the infarct border zone in rabbit myocardium and in mice harboring an red fluorescent protein-green fluorescent protein-LC3 transgene. In cultured myocytes subjected to I/R, SAHA overtly alleviated cell death, the effect of which was correlated with increased autophagy. The permissive role of autophagy in SAHA-related beneficial effects was consolidated by the mitigation of SAHA efficacy through RNAi knockdown of autophagy genes Atg7 and Atg5. These findings have great clinical relevance because the plasma SAHA levels were similar to those achieved in cancer patients. This work has unveiled a new paradigm for the clinical utility of HDAC inhibitors and autophagy regulators in ischemic heart diseases. A plethora of studies have demonstrated proven cardioprotective benefits of HDAC inhibitors in models of myocardial stress, including cardiac hypertrophy, I/R, and heart failure.

In particular, trichostatin A, a class I and II HDAC inhibitor structurally homologous to SAHA, reduces myocardial infarct size up to 50%. HDAC inhibition caused a dramatic increase in phosphorylation of p38 and p38 activity in the heart. Of note, HDAC inhibitors can be delivered as late as 1 hour after an ischemic insult and can achieve a similar degree of infarct size reduction using pretreatment, indicating the suitability of HDAC inhibitors to treat myocardial infarction at the time of percutaneous coronary intervention. Although discrepancies exist in disease mechanisms in animal models relative to the human case, these data clearly show that facilitated autophagy is required for HDAC inhibition–induced protection against I/R injury. Given the recent therapeutic promises using HDAC inhibitors in ischemic and hypertrophic heart diseases, the finding that SAHA rescues I/R heart injury through modulating autophagy flux is of great clinical importance. Interestingly, a number of cardioprotective agents such as the angiotensin II receptor blocker valsartan may also elicit protection against I/R injury through autophagy induction. Valsartan preconditioning is believed to facilitate autophagy induction via an Akt/mammalian target of rapamycin/6K–mediated mechanism, although the underlying molecular mechanism behind SAHA-induced autophagy flux remains unclear at this time.
fusion. It has been demonstrated that Atg4 is subject to oxidation, thus resulting in LC3-II accumulation and autophagosome formation. I/R injury–induced ROS production has been demonstrated to promote nuclear factor-xB activity and to accentuate myocardial injury via the activation of Beclin-1–mediated autophagy. On the other side of the coin, a number of autophagy regulatory proteins such as the mitochondrial autophagy protein Parkin possess multiple conserved cysteine residues that are susceptible to modification by oxidative and nitrosative stress, leading to aberrant modification and subsequently inhibition of autophagy. Considering the current controversy on the role of adaptive or maladaptive autophagy in cardiac pathology, it is pertinent to better understand conditions in which autophagy should be inhibited or activated not only to best preserve cardiac homeostasis but also to optimize drug therapy (such as the drug efficacy and resistance for HDAC inhibitors). Furthermore, it is noteworthy that the overall efficaciousness of HDAC inhibition is determined by the pleiotropic processes, many of which (eg, anti-inflammation and NADPH oxidase) may be autophagy independent.

HDACs are pivotal epigenetic regulatory enzymes with the ability to deacetylate nucleosome histones and nonhistone proteins and to elicit significant pathological effects in tumor growth and cardiovascular diseases. Two HDAC inhibitors, vorinostat and romidespin, have been approved by the Food and Drug Administration for cancer therapy. Although small-molecule HDAC inhibitors have displayed an unforeseen potential in the management of heart diseases, the broad-spectrum, “pan” HDAC inhibition is associated with toxicities such as thrombocytopenia, neutropenia, diarrhea, nausea, vomiting, and fatigue. Experts in the field remain skeptical of the prospects of translating these preclinical findings to the clinical setting. With the improved safety profiles of newly identified small-molecule HDAC inhibitors selectively targeting 1 or a small subset of the 18 human HDACs, identifying the HDAC isoform(s) that govern pathological cardiac remodeling and understanding the roles of acetylation/deacetylation in the regulation of autophagy and autophagy-independent cellular processes and the underlying molecular mechanisms should be the logical next steps for the advancement of safer HDAC inhibitors into clinical trials for heart diseases, particularly ischemic heart disease.

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Disclosures

None.

References

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