HDAC Inhibition in the Heart
Erasing Hidden Fibrosis

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Altering the transcriptional profile of cells govern downstream events fundamental to both physiology and pathology. Central in this control of transcription are changes in chromatin structure elicited by post-translational modifications of histone proteins.1 The protein machinery that act to add, remove, or detect these modifications are known colloquially as writers, erasers, and readers of histone modifications.2 There is great interest in the clinical application of small molecules that alter the activity of these proteins in the hope of fostering clinical benefit. Small molecule inhibitors of a class of “erasers” known as histone deacetylases (HDACs) have made their way to the clinic.3

As the name implies, HDAC proteins remove acetyl groups from histone protein tails, but they can also target nonhistone proteins. HDACs fall into 4 different classes (I, II, III, and IV). Class I, II, and IV share a conserved zinc-dependent functional domain and most small molecule inhibitors in use target that domain.3 A number of HDAC inhibitors (HDACis) have been approved by the US Food and Drug Administration for the treatment of various cancers, with >60 clinical trials reported as currently active on clinicaltrials.gov. The list of approved drugs includes the pan-HDACis vorinostat, panobinostat, and belinostat, which inhibit class I, II, and IV HDACs, and the more specific class I HDACi romidepsin.3 The safety and efficacy of these drugs in treating cancers have spurred preclinical testing for use in a variety of other diseases including heart disease.2,3

Early indications of the potential of HDACi to treat heart disease emerged from results using a mouse model of aortic constriction.4,5 This model of pressure overload–induced hypertrophy is marked by development of pathologic ventricular hypertrophy progressively leading to cardiac dilation and emergence of the phenotype of heart failure with reduced ejection fraction. Treatment with a pan-HDACi resulted in a blunting of the hypertrophic response and a slowing of progression to failure.4,5

Markers of cardiac fibrosis were also decreased in the ventricle, and HDACi blunted collagen production in isolated cardiac fibroblasts.5 HDACi was also capable of promoting regression of established hypertrophy.4 These fundamental cardioprotective findings have been replicated in a number of studies, including a study using cardiomyocyte-specific inactivation of the class I HDACs (HDAC1 and HDAC2).2,6 In contrast, genetic studies using cardiomyocyte-specific deletions of specific class II HDACs reported an enhanced hypertrophic response.7 These opposing data suggest that class I HDAC activity may dominate class II–dependent events in the response to hypertrophic stress.

HDACi can protect the ischemic heart both by limiting the extent of cell death at reperfusion and by reducing the pathologic remodeling that occurs subsequently. In both a murine and rabbit model of ischemia/reperfusion, a single dose of HDACi administered...
at the time of reperfusion reduced the size of the resulting myocardial infarction by as much as 50%. Pathologic remodeling in response to ischemic injury is also inhibited by HDACi. Both fibrosis and cardiomyocyte hypertrophy at the infarct border and remote zones of the heart are reduced by HDACi. HDAC inhibition has been shown to target the inflammatory cascade that accompanies heart failure.

Heart failure with preserved ejection fraction (HFpEF) is a syndrome growing in prevalence with no effective therapies available. It is characterized by LV diastolic dysfunction, and a number of studies have begun to examine whether HDACi is effective in preventing or reversing models of diastolic dysfunction. One study using the new clinical-stage HDACi givinostat demonstrated that HDACi can improve cardiac performance in 2 different models of diastolic dysfunction: the hypertension-induced Dahl salt-sensitive rat and a model of aging-induced diastolic dysfunction in normotensive mice. HDACi-induced improvement in these models appeared to be independent of blood pressure, cardiac hypertrophy, or changes in cardiac sarcomeric protein isoform expression, as well as regulation of gene transcription. Instead, improvement was correlated with enhancement of cardiac myofibril relaxation through direct deacetylation of sarcomeric proteins. Working with isolated cardiac myofibrils, the investigators revealed impairment of cardiac myofibril relaxation that could be overcome by directly decreasing the acetylation levels of the sarcomeric proteins. A recent article from some of the same authors using a feline model of pressure overload–induced diastolic dysfunction and heart failure showed a similar effect of vorinostat on isolated cardiac myofibril relaxation. In the latter study, HDACi also promoted decreases in cardiac hypertrophy and cardiac fibrosis similar to those reported in mouse models of pressure overload–induced hypertrophy.

A new study by Travers et al in this issue of Circulation used a uninephrectomy plus deoxycorticosterone acetate–salt (UNX/DOCA) hypertensive mouse model of diastolic dysfunction to expand on earlier findings. The investigators demonstrate that HDAC inhibition using givinostat reverses established diastolic dysfunction in this model. Four weeks after UNX/DOCA, the animals developed diastolic dysfunction as suggested by a reduction in mitral Doppler E/A and an increase in E′/E′ ratios assessed by serial echocardiography, as well as elevated LV end diastolic pressure as measured by invasive hemodynamic analysis. Within 2 weeks of HDACi treatment, the hearts demonstrated a nearly complete recovery of these measures of diastolic dysfunction with no changes in DOCA-induced hypertrophy.

This finding on its own is not unexpected. What is surprising is that unlike the earlier studies, the authors observed no impairment of cardiac myofibril relaxation in myofibrils isolated from the UNX/DOCA model, nor were changes in contractile performance in intact myocytes, including maximal rates and time constants of contraction and relengthening, observed in this model. Instead, by using quantitative mass spectrometry the investigators uncovered what they term “hidden LV fibrosis” in the UNX/DOCA-treated animals. This phenomenon is heralded by a significant increase in collagen levels detected in the extracellular matrix (ECM) at a point where detection of fibrosis by traditional means revealed no differences. These changes in ECM peptides correlated not only with the observed diastolic dysfunction, but also with an increase in a measurement of LV tissue “stiffness” detected by atomic force microscopy. Treatment of the animals with givinostat reverted the ECM peptide profile back to control levels as well as the metrics of LV stiffness. These changes correlated with an improvement in diastolic function. The investigators went on to show that givinostat can inhibit transforming growth factor–β activation in fibroblasts by decreasing the localization to target genes of the histone acetylation “reader” protein BRD4 (bromodomain-containing protein 4). This reader/eraser paradox has also been observed in cardiac hypertrophy, where BRD4 binding is required for the expression of hypertrophic genes and yet HDACi that increase acetylation on histones (and presumably BRD4 association with chromatin) decrease expression of hypertrophic genes. The simple answer to this seeming paradox may lie in recognizing that this biology is highly dynamic, with acetylation and deacetylation allowing release and binding of BRD4 for proper recruitment and activation.

HDAC inhibition has the potential to treat a wide range of disease states. The specific transcriptional pathways affected by HDACi are likely influenced not only by the unique chromatin architecture across cell types but also by changes in chromosome structure in response to the microenvironment to which the cells are exposed. When it comes to heart disease, however, some themes have emerged. HDACi appears to inhibit cardiac hypertrophy, protect against oxidative damage, inhibit inflammation, inhibit fibrosis, and modulate ECM composition (Figure). The detection of this “hidden fibrosis” is especially intriguing and raises some interesting questions: How universal are these changes, and can they be observed in samples from patients with HFpEF? That diastolic dysfunction is ameliorated independent of cardiomyocyte “stiffness” suggests that the majority of cardiac stiffness in this hypertensive setting lies in the ECM and fibrosis rather than the cardiomyocytes per se. Hence, more accurate methods (eg, magnetic resonance imaging) of detecting early, minimal cardiac fibrosis may be warranted in the evaluation of patients with hypertension at risk of developing HFpEF. ECM not only plays a structural role but can also affect cell signaling, cell growth, and recruitment of inflammatory cells. What is the effect of these ECM changes in the development of HFpEF? These studies highlight the potential for HDACi in the treatment of metabolic HFpEF, where inflammation plays a key role. Although still in the preclinical...
phase of development, the future of HDACi in the treatment of cardiac disease is not hidden.

**ARTICLE INFORMATION**

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**Disclosures**

Dr Gillette is a coinventor on a patent application (number PCT/US/2017/037019) that was filed in June 2017 (provisional application filed in June 2016). The patent relates to the diet used for modeling heart failure with preserved ejection fraction.

**REFERENCES**


