In general clinical terms, heart failure (HF) is manifested by defects in cardiac pump function (ejection, filling, or a combination of both) that, in turn, will cause clinical signs and symptoms that are often progressive and give rise to exercise limitations. Heart failure (HF) is a complex clinical syndrome that is typically characterized by left ventricular (LV) systolic dysfunction, with a reduced ejection fraction (EF) or preserved EF. The morbidity and mortality associated with HF have been increasing, and it is estimated that there are approximately 5.7 million people in the United States with HF, representing a significant public health burden. The pathophysiology of HF is complex and involves multiple factors, including myocardial hypertrophy, remodeling, and fibrosis. The role of the myocardial matrix in the development and progression of HF is now recognized as a key component in the pathophysiology of the disease. The myocardial matrix plays a critical role in the maintenance of myocardial function and structure, but its dysfunction can lead to the development of HF.

**Abstract**—In contrast to public perception, the morbidity and mortality and the resultant healthcare costs associated with chronic heart failure (HF) are increasing and arguably reaching epidemic proportions. Although basic research efforts have provided unique insights into fundamental processes that govern myocardial extracellular matrix (ECM) growth and function, the translation of these findings to improved diagnostics and therapeutics for HF has not been as forthcoming. The factors that contribute to this relative paucity of new clinical tools for HF are multifactorial but likely include the need to recognize and differentiate HF phenotypes and to couple the use of biomarkers and multimodality imaging in early translational research studies. Recognizing the classification scheme of HF with a reduced ejection fraction (EF) to that of HF with a preserved EF and incorporating unique and differential measurements of ECM remodeling to these specific disease processes are warranted. For example, profiling pathways of ECM degradation such as the matrix metalloproteinases in patients with ischemic heart disease and HF with a reduced EF can provide prognostic information in terms of risk of progression to HF. In patients with chronic hypertensive disease and HF with a preserved EF, plasma profiling indexes of ECM synthesis and turnover, as well as advances in ECM imaging, have been shown to provide diagnostic and prognostic use. In terms of therapeutics, strategies to stabilize the ECM in HF with a reduced EF hold potential, whereas in contradistinction, selective antifibrotic agents may hold promise for HF with a preserved EF.

**Clinical Trial Registration**—URL: http://www.clinicaltrials.gov. Unique identifier: NCT 00946231

**Key Words:** cardiovascular diseases ■ diagnostics ■ myocardial remodeling ■ therapeutics ■ ventricular function
LV afterload such as hypertension or aortic valve disease

In contrast, HF that arises from an increase in long-standing ischemic heart disease would likely give rise to HFrEF.4,6,9

significant myocardial injury and loss of contractile units such as ischemic injury and MI first occurs through the classic cell death pathway, necrosis. This region of necrotic myocytes then causes a cascade of biological events that can be considered a wound-healing process, and thus the canonical phases of wound healing are often used, which consist of 3 overlapping phases: inflammation, proliferation, and maturation. The first phase is the acute period in which reactive oxygen species, bioactive signaling molecules, and peptides released from the local environment cause inflammatory cell recruitment and invasion. The initiation of the inflammatory phase is likely attributable to the release of molecules as a result of myocyte death and, in turn, induces the expression of cytokines such as tumor necrosis factor-α and the interleukins. A more comprehensive review of this particular subject was provided in a recent issue of Circulation Research.11 Some key aspects of the early post-MI inflammatory process include (1) activation of the intracellular transduction pathway, nuclear factor-κB; (2) induction of a diverse family of chemokines; and (3) neutrophil recruitment and margination. These cytokines and signaling pathways likely contribute to the movement and expansion of monocytes and ultimately the maturation into macrophages within the MI region. Although initially considered a rather monochromatic cell type in terms of form and function, the macrophages within the MI region seem to be quite heterogeneous and may ultimately influence the magnitude and direction of the localized inflammatory process.11–13 Although this inflammatory process is critical to initiate a reparative response after myocardial injury, it seems that, unlike other sites of tissue injury, the inflammatory response is prolonged within the MI region. The second phase is heralded by proliferation and transdifferentiation of myocardial fibroblasts into myofibroblasts.14–17 Furthermore, released matrix structural proteins such as fibrillar collagens and matrix-integrin-cytoskeletal interactions facilitate contraction of the wound. The third phase of the wound-healing process normally results in complete contraction of the wound, apoptosis of the myofibroblasts, and the formation of a relatively acellular scar. However, in the context of post-MI remodeling, this canonical set of events does not necessarily occur. Rather, there is a continued proliferation and transdifferentiation of myofibroblasts within the MI and border regions accompanied by a shift in matrix proliferative/degradative pathways within these transformed cells. A schematic that summarizes these cellular/ECM events is shown in Figure 1.
The affected region after the MI that contributes to infarct expansion, progressive LV remodeling, and systolic dysfunction not only is composed of the MI region itself but also can affect the viable myocardium surrounding the MI (border zone), as well as the remote, normally perfused region of the LV. The remote, viable myocardium is subjected to changes in radial wall stress as a function of the hypokinetic, akinetic, or dyskinetic region that encompasses the MI and infarct expansion zone. This increased wall stress evokes a hypertrophic response that shares some characteristics that occur with pressure-overload hypertrophy, and the physiological consequences of this process are best exemplified in the section on HFrEF.

Significant changes within the ECM occur during all time points after MI and likely contribute to the overall adverse LV remodeling process. First, the inflammatory response causes the release of matrix metalloproteinases (MMPs) and other proteases to degrade the ECM and to allow margination of inflammatory cells. However, with a persistent inflammatory state, MMP induction will also destabilize the newly formed ECM and the nascent scar. Second, the continued proliferation and altered expression of the myofibroblasts within the MI region in terms of MMP and tissue inhibitor of MMPs (TIMP) expression will cause ECM instability and infarct expansion. Specifically, this transformed fibroblast population within the MI region likely results in a shift in the relative balance of MMPs and TIMPs, favoring accelerated ECM turnover and a failure of mature scar formation. Third, although a robust expression of profibrotic signaling molecules occurs within the myofibroblasts and residual myocyte populations such as transforming growth factor (TGF)-β, this does not result in a well-formed ECM. For example, although TGF signaling is enhanced within the MI region and causes significantly higher fibrillar collagen transcription, several critical posttranslational events involving collagen assembly may be defective. For example, a loss of the matricellular protein secreted protein acidic and rich in cysteine, which is involved in procollagen processing to a mature collagen fibril, resulted in impaired collagen maturation and MI scar formation. However, although fibrillar collagens have formed the focus of early basic and clinical research on post-MI remodeling, the ECM is a highly diverse entity, and several noncollagen structural and nonstructural proteins likely play a critical role in this process. For example, the interactions of the glycoproteins, fibronectin, and tenascin C may directly alter cell-ECM interactions, whereby other bioactive molecules such as osteopontin can directly influence ECM biosynthesis through several signaling pathways. Transmembrane receptors and coreceptors such as endoglin and syndecan that can modify profibrotic signaling through TGF and other growth factors also likely play an important role not only in ECM synthesis and degradation but also in myofibroblast transformation. Thus, abnormalities in fibroblast phenotype coupled by defects in both ECM synthesis and degradation pathways likely converge to cause a feed-forward process in terms of MI wall thinning and dyskinesis, increased radial wall stress, and local strain patterns and ultimately contribute to overall infarct expansion, LV remodeling, and HFrEF. These pathways and processes, as they may relate to the prognosis and diagnosis of HFrEF, are presented in a subsequent section.
Current Diagnosis and Treatment

Although several pharmacological/device-driven therapies are used with HFrEF secondary to MI, it should be emphasized that these interventions are considered for managing symptoms and attenuating the relative progression of LV pump dysfunction. Although these treatment modalities can potentially slow the progression and severity of HFrEF, these approaches do not necessarily stop or reverse the underlying LV remodeling process that continues in an inexorable fashion. Relevant to the focus of this review, several past studies have demonstrated a direct relationship between the rate and progression of this adverse LV remodeling process after MI and specific determinants of ECM remodeling. Thus, as discussed in a subsequent section, an important research direction is to develop diagnostic and therapeutic strategies that target specific underling mechanisms such as those that occur within the ECM that result in adverse LV remodeling after MI.

HFrEF: The Pressure-Overloaded Myocardium

The fundamental milestone in the development and progression of HFrEF is concentric LV hypertrophy, which is a result of accelerated growth of both the myocyte and the ECM compartments. The stimulus for LV hypertrophy is a prolonged pressure overload that can arise as a function of hypertension, aortic stenosis, and arterial stiffening as a function of age. Although LV hypertrophy is initially an appropriate adaptive response, continued myocyte growth and expansion of the LV will eventually lead to fundamental defects in LV diastolic performance. As illustrated in Figure 1, increased fibrillar collagen accumulation and fibroblast/myofibroblast proliferation within the ECM in the context of HFrEF are common features. Thus, although defects in systolic function can certainly be operative in severe LV hypertrophy and increased afterload, the key pathophysiological features of HFrEF are increased LV mass and impaired diastolic function. In general terms, LV diastolic function is determined by 2 fundamental processes: active relaxation and passive stiffness. In terms of active relaxation, this is highly dependent on removal of Ca\textsuperscript{2+} from the myofilaments, allowing a decrease in the cross-bridge cycling rate and a return to resting sarcomere lengths. With significant LV myocyte hypertrophy, a not-uncommon finding is that of impairments in Ca\textsuperscript{2+} handling and thus slowing of the return to resting sarcomere length. Changes in cytoskeletal structure and function also occur with myocyte hypertrophy, which can include altered expression and function of the high-molecular-weight proteins. For example, the high-molecular-weight protein titin may be altered in terms of isoform type and phosphorylation state with LV hypertrophy, which may include aggressive medication strategies for hypertension and aortic valve replacement for aortic valve stenosis. However, removal/reduction of the LV pressure-overload stimulus such as the treatment of high blood pressure is insufficient. For example, concentric LV hypertrophy and diastolic dysfunction commonly develop without concomitant symptoms and often before the presence of significant hypertension has been detected. In addition, despite rigorous blood pressure control and removal of the afterload stimulus, LV hypertrophy and progressive diastolic dysfunction can occur, particularly ECM accumulation. Thus, an important translational research direction is to develop diagnostic strategies that can detect early changes in the LV remodeling process in patients at increased risk for HFrEF, as well as methods by which to effectively monitor this process over time. Accordingly, integrating measures of ECM remodeling may provide important insight into the natural history of HFrEF and may provide prognostic information in terms of the risk for progression/acceleration of LV stiffness and attendant symptoms of HF.

Comparisons between the relative effectiveness of what is considered to be standard of care for patients with HFrEF and
that of patients with HFrEF underscores that distinct differences in the cellular/extracellular basis of the HF disease process likely exist.\textsuperscript{8-10} For example, when angiotensin receptor blockers were used in robust samples of patients with HFrEF,\textsuperscript{2,30,31} no significant treatment effect on a primary composite response variable was obtained. These clinical studies of HFrEF emphasize the significant differences in the pathophysiology of this disease process and the relative refractoriness of the LV remodeling and dysfunction that occur when significant symptoms of HF become manifest in the context of HFrEF. Indeed, once HFrEF has developed, subsequent 5-year morbidity and mortality rates approach or exceed 60%, and 6-month rehospitalization rates are 50%.\textsuperscript{2,4,6,7} Furthermore, once LV remodeling has developed such as ECM remodeling and HFrEF, even the presence of Seventh Report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure recommended blood pressure control does not reduce morbidity and mortality. Thus, in light of the fact that changes in ECM synthesis/degradation play a critical role in LV myocardial stiffness, which is a fundamental determinant of the HFrEF process, defining mechanistic pathways and therapeutic targets that are uniquely targeted to the ECM and the progression to HFrEF constitutes an important and clinically relevant research focus.

**Future Directions for HFrEF and HFrEF: Diagnostics and Therapeutics**

With the development of HF, significant LV remodeling and dysfunction have occurred; thus, treatment is often palliative. Therefore, an important research direction is to develop diagnostic strategies that can be used in large patient populations that would identify early onset of disease progression. One potential diagnostic approach is the measurement of proteins, bioactive molecules, and microRNAs (miRs) in a peripheral blood sample, which may indicate the presence and the relative progression of the HF process. The generic term for these measurements is biomarkers, and biomarker profiling may be a useful strategy for the detection, identification, differentiation, and evaluation of therapeutic efficacy in the context of HF. This is a rapidly growing field, with >700 publications in 2012 alone containing the key words of HF (PubMed, Search, March 13, 2013). Some examples of research directions and potential applications of biomarker profiling as it applies to HF with a specific focus to determinants of the ECM are presented below. However, it should be recognized that biomarker signatures can also apply to indexes of LV myocardial remodeling through imaging approaches. This is also a rapidly evolving field, and translational/clinical research examples of this approach in terms of potential diagnostic/prognostic strategies are briefly presented.

**Diagnostics and HFrEF**

In terms of plasma biomarker profiling and a post-MI pathogenesis, plasma troponin levels are relevant.\textsuperscript{24} There are several important lessons from the use of plasma troponin. First, the use of this biomarker to definitively identify an acute coronary syndrome and myocardial injury has progressed to become a point-of-care test. Thus, although some biomarker measurements currently require complex immunoassays with a significant delay in results, it is highly likely that more high-throughput results with clinically relevant turn-around times will be developed. Second, although plasma troponin values were initially used for the index event, high-sensitivity troponin measurements have identified the presence of ongoing myocyte injury in patients with HFrEF and hold prognostic potential. Third, the use of high-sensitivity troponin along with other biomarkers such as N-terminal brain natriuretic peptide (BNP) and indexes of inflammation provided additive predictive value in terms of outcomes in patients with HFrEF.\textsuperscript{32} Because the activation of several inflammatory cascades occurs after MI and can directly influence ECM remodeling pathways, it follows that cytokine profiling holds relevance in terms of diagnostic and prognostic information with HFrEF.

Indeed, a past study using a multivariable modeling algorithm not only exemplified the incremental and additive effects of profiling cytokines of different domains but also underscored the diminishing return of broadband biomarker measurements.\textsuperscript{33} Thus, it is likely that a specific portfolio of biomarkers will provide specific diagnostic information in the early post-MI period, whereas a different biomarker panel may be of use in later post-MI time points. This concept is becoming realized in terms of using plasma biomarkers of ECM remodeling in patients after MI, as detailed in Table 1. For example, early studies have demonstrated that increased plasma levels of collagen peptide fragments occur after MI and in progressive HFrEF.\textsuperscript{32,34-36} In keeping with ECM degradation, induction of ECM proteases such as the MMPs occurs in patients after MI and likely holds predictive value in terms of identifying those patients at greatest risk for the early development of HFrEF.\textsuperscript{32,36-41} For example, the magnitude of plasma MMP-9 levels that occur within the early post-MI period is associated with a greater degree of LV dilation at 6 months after MI.\textsuperscript{38} In a recent report that compiled the results from >60 studies in patients after MI, those that were most consistently present in prediction models included indexes of neurohormonal activation such as BNP and indexes of ECM degradation such as MMP-9.\textsuperscript{36} Cytokine/chemokine release, which would cause the induction of pathways relevant to ECM degradation and myofibroblast transformation, and the release of matrixines occur in patients in the post-MI period.\textsuperscript{32,42-45} For example, release of soluble cytokine receptors such as interleukin-6 and soluble ST2, which in turn likely induce several ECM remodeling pathways, is associated with disease progression in HFrEF.\textsuperscript{32,42} Other matrix-related molecules such as plasma galectin-3 have been associated with other determinants of ECM remodeling but not necessarily independently predictive of HFrEF.\textsuperscript{46-48} In addition to plasma biomarkers for ECM degradation, increased markers for ECM synthesis have been identified, which include the TGF coreceptor endoglin and the matrixines tenascin-C, osteopontin, and syndecan-4.\textsuperscript{33,49-53} In addition to profiling downstream indexes of myocardial viability, inflammation, and ECM remodeling in patients at risk for HFrEF, it may be possible to use peripheral blood sampling to monitor upstream regulatory processes at the posttranscriptional level through quantifying miRs. These miRs directly influence posttranscriptional events through several molecular interactions, which include binding to mRNA and interfering with initiation of translation and accelerating...
mRNA degradation. Although an area of active research and a focus of a recent review series in Circulation Research, it is becoming apparent that specific miRs will cause posttranscriptional alterations in functional clusters of proteins that would be relevant to LV remodeling and, in particular, to the ECM. For example, in a mouse model of MI, changes in relative myocardial miR-29a levels had pronounced and directional effects on ECM remodeling in terms of myocardial fibrosis. Robust Table 1. Matrix Biomarker Profiling in HFrEF

<table>
<thead>
<tr>
<th>Biomarker(s)</th>
<th>Primary Observation</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>PINP, ICTP, PIINP, MMP-1, TIMP-1</td>
<td>Longitudinal changes in all ECM markers whereby ICTP is associated with greater HF symptom progression</td>
<td>34</td>
</tr>
<tr>
<td>PIINP, MMP-1, TIMP-1, hsCRP, IL-18, IL-10</td>
<td>Indexes of increased ECM turnover associated with functional capacity and outcomes</td>
<td>35</td>
</tr>
<tr>
<td>MMP-1, TIMP-1, and biopsy</td>
<td>Shift in MMP-1/TIMP-1 balance favoring ECM degradation as evidenced by ECM biopsy histology</td>
<td>26</td>
</tr>
<tr>
<td>52 Biomarkers evaluated including determinants of ECM remodeling</td>
<td>Biomarkers most consistently associated with LV remodeling were involved in ECM turnover such as collagen pro- and telo-peptides and MMP-9</td>
<td>36</td>
</tr>
<tr>
<td>MMP-2, MMP-9, hsCRP, TNF-α, pro-BNP</td>
<td>Higher plasma MMP-9 levels associated with HF progression</td>
<td>37</td>
</tr>
<tr>
<td>Portfolio of MMPs and TIMPs serially examined</td>
<td>A specific cassette of MMPs is increased in plasma after MI and associated with progressive LV remodeling</td>
<td>38</td>
</tr>
<tr>
<td>N-BNP, MMP-2, MMP-9</td>
<td>≤4 y of follow-up, plasma MMP-9 levels associated with magnitude of LV systolic failure</td>
<td>39</td>
</tr>
<tr>
<td>TIMP-1, -2, -4</td>
<td>Early changes in plasma TIMP-4 levels associated with degree of LV dilation at 3 mo after MI</td>
<td>40</td>
</tr>
<tr>
<td>TIMP-1, MMP-9, NT-proBNP</td>
<td>Changes in MMP-9 associated with adverse post-MI remodeling as defined by LV dilation and TIMP-1 levels demonstrated predictive risk for combined event end point (HF/death)</td>
<td>41</td>
</tr>
<tr>
<td>sST2, hsTnT, NT-proBNP</td>
<td>Combining biomarkers from different functional domains improved prognostic potential for progression to HF</td>
<td>32</td>
</tr>
<tr>
<td>CCL19, CCL21</td>
<td>Increased CCL21 associated with mortality</td>
<td>43</td>
</tr>
<tr>
<td>gp130, IL-6</td>
<td>Increased gp130 associated with all-cause and cardiovascular-related mortality and progressive HF</td>
<td>44</td>
</tr>
<tr>
<td>G-CSF, MCP-1, M-CSF</td>
<td>M-CSF-1 associated with increased risk of all-cause mortality</td>
<td>44</td>
</tr>
<tr>
<td>TNF, IL-6, and respective soluble receptors</td>
<td>Demonstrated power of time-series ensemble models to improve predictive accuracy of HF outcomes</td>
<td>42</td>
</tr>
<tr>
<td>GAL-3</td>
<td>GAL-3 affected by renal function but not by presence or absence of HF progression</td>
<td>46</td>
</tr>
<tr>
<td>100, 1562</td>
<td>GAL-3 not associated with HF progression or outcomes but correlated with MMP-3</td>
<td>47, 48</td>
</tr>
<tr>
<td>Soluble endoglin</td>
<td>Increased s-endoglin associated with major cardiovascular events, including progressive HF</td>
<td>49</td>
</tr>
<tr>
<td>TN-C, BNP</td>
<td>Early changes in TN-C levels after MI associated with HF</td>
<td>33</td>
</tr>
<tr>
<td>OPN</td>
<td>Increased early OPN levels after MI associated with all-cause mortality and cardiovascular outcomes</td>
<td>50</td>
</tr>
<tr>
<td>OPN</td>
<td>OPN may provide significant prognostic information independently of other traditional prognostic markers in patients with stable IHD</td>
<td>51</td>
</tr>
<tr>
<td>OPN</td>
<td>OPN associated with progressive HF</td>
<td>52</td>
</tr>
<tr>
<td>Syndecan-4</td>
<td>Increased syndecan-4 associated with poor LV systolic function</td>
<td>53</td>
</tr>
</tbody>
</table>

BNP indicates B-type natriuretic peptide; CCL, chemokine ligand; ECM, extracellular matrix; GAL-3, galectin-3; G-CSF, granulocyte colony–stimulating factor; gp130, glycoprotein 130β; HF, heart failure; HF/EF, HF with reduced ejection fraction; hsCRP, high-sensitivity C-reactive protein; hsTnT, high-sensitivity troponin T; ICTP, carboxy-terminal telopeptide of collagen I; IHD, ischemic heart disease; IL, interleukin; LV, left ventricular; MCP-1, monocyte chemotactic protein-1; M-CSF, macrophage colony–stimulating factor; MI, myocardial infarction; MMP, matrix metalloproteinase; NT-proBNP, N-terminal pro-brain natriuretic peptide; OPN, osteopontin; PIINP, procollagen III N-terminal propeptide; PINP, procollagen type I N-terminal propeptide; pro-BNP, pro-brain natriuretic peptide; TIMP, tissue inhibitor of MMPs; TN-C, tenascin-C; and TNF, tumor necrosis factor.

*Not intended to be comprehensive but rather representative of studies using peripheral blood sampling.
and early changes in relative plasma levels of miR-29a have been identified in patients after MI, as shown in Figure 2.57 More important, this past study demonstrated that early changes in plasma miR-29a levels were associated with adverse LV remodeling at 3 months after MI. This is an example of what is likely to be a large cluster of miRs that are altered after MI and may hold prognostic use in terms of identifying those patients at increased risk for HFpEF, as well as potentially monitoring the progression of this disease process. Thus, future translational research that couples basic mechanistic studies of miR function and specific ECM-related targets to that of peripheral profiles of miRs would certainly be of great relevance.

Cardiac imaging with ultrasound, magnetic resonance imaging (MRI), or x-ray computed axial tomography provides critical information on LV geometry and function after MI and assesses the severity of the HFpEF process. However, research is moving beyond these basic imaging measures and incorporating assessment of molecular signatures that may underlie the fundamental disease process itself.38–41 Specifically, in animal studies using single photon emission computed tomography with specifically labeled isotopes, it has been possible to identify changes in receptor signaling pathways, inflammation, and MMP activation after MI. As an example, with the use of a technetium-99m–labeled angiotensin receptor blocker (99mTc-losartan), time-dependent changes in angiotensin receptor density were identified in a post-MI murine model.59 In addition, assessment of myocardial sympathetic activation with metaiodobenzylguanidine perfusion defect at the site of the MI was evident (top, arrow) and intense MMP activity could be identified in a post-MI pig model.60 The next research steps will be to incorporate these imaging modalities to provide quantification of LV geometry and function, as well as molecular signals that may initiate and promulgate the adverse LV remodeling process after MI.

Diagnostics and HFpEF

Although arguably an imprecise term, myocardial fibrosis has been assigned the definition for the increased ECM that invariably accompanies HFpEF and the attendant changes in myocardial stiffness. Because the ECM plays a critical role in the pathophysiology of HFpEF, strategies that can evaluate and quantify changes in this entity remain an important research direction. A summary of those plasma biomarkers that have been evaluated in the context of HFpEF that may hold relevance to ECM remodeling is presented in Table 2. For example, plasma profiles of peptide fragments of fibrillar collagens (types I and III) have been used in patients with developing and established HFpEF.27,28,39,62,63 The collagen fibril may undergo posttranslational modification by enzyme-mediated cross-link reactions and non–enzyme-mediated cross-link formation. Of note, non–enzymatic cross-link formation can be mediated by advanced glycation end products (AGEs).64,65 AGEs bind to specific cell membrane receptors that include receptor for AGE. The extracellular ligand binding domain of the receptor, soluble receptor for AGE, is released into the ECM and thus can be released into the vascular compartment. Because increased fibrillar collagen stability can occur through increased receptor for AGE interactions, profiling plasma levels of soluble receptor for AGE and indexes of AGE formation may hold prognostic/functional relevance in HFpEF.64,65 A biologically relevant consequence of increased collagen AGEs is reduced susceptibility to MMP-mediated degradation and turnover. In parallel, significant and directional changes in the plasma profiles of MMPs and TIMPs occur in patients with developing HFpEF and have been shown to hold potential prognostic use.66–69 For example, a specific cassette
of MMPs and TIMPs, coupled with N-terminal BNP, provided significant prediction in terms of patients at risk for development of HFpEF (Figure 4).66 In this past study, it was demonstrated that profiling-specific MMPs and TIMPs provided significant and additive predictive value compared with N-terminal BNP measurements alone and that this multimarker predictive model withstood cross-validation analysis.66 Other ECM chemokines/matrikines such as cardiotrophin have been associated with the development and progression of HFpEF.70–74

Table 2. Matrix Biomarker Profiling in HFpEF*

<table>
<thead>
<tr>
<th>Subjects, n</th>
<th>Biomarker(s)</th>
<th>Primary Observation</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Matrix metabolism</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>156</td>
<td>MMP-1, TIMP-1, PINP</td>
<td>TIMP-1 levels were highest in patients with elevated PCWP, whereas relative MMP-1 levels were lower and PINP levels were higher</td>
<td>28</td>
</tr>
<tr>
<td>171</td>
<td>PICP, PINP, CTIP, PIINP, MMP-1, MMP-2, MMP-9, CTX, serum carboxy-terminal, amino-terminal</td>
<td>A combinatorial predictive model of collagen peptides and MMP-2 levels in patients with indexes of worsening diastolic function</td>
<td>38, 75</td>
</tr>
<tr>
<td>880</td>
<td>PICP, ICTP, PIINP</td>
<td>PICP and ICTP elevated in elderly patients with HF</td>
<td>62</td>
</tr>
<tr>
<td>334</td>
<td>PINP, PIINP, OPN</td>
<td>Stepped increases in procollagen and osteopontin caused incremental increases in the hazard ratio for cardiovascular events</td>
<td>63</td>
</tr>
<tr>
<td>580</td>
<td>AGEs, sRAGE</td>
<td>Indexes of AGEs such as pentosidine and soluble receptor for AGE increased in plasma in HFpEF and in univariate models associated with clinical outcomes</td>
<td>64</td>
</tr>
<tr>
<td>880</td>
<td>PICP, ICTP, PIINP</td>
<td>CTP specifically associated with worsening HF and hospitalization in patients with HFpEF but not HFrEF</td>
<td>29</td>
</tr>
<tr>
<td>26</td>
<td>AGEs</td>
<td>Indexes of increased AGEs associated with diastolic dysfunction</td>
<td>65</td>
</tr>
<tr>
<td><strong>Matrix protease</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>446</td>
<td>MMP-1, -2, -3, -7, -8, -9, TIMP-1, -2, -3, -4, NT-proBNP, OPN, sRAGE, ICTP, PIINP</td>
<td>Biomarkers reflective of ECM synthesis (ie, PIINP) and modified degradation rates (ie, TIMP-4) provided additive predictive value in a multivariable model to predict presence and severity of HF</td>
<td>66</td>
</tr>
<tr>
<td>103</td>
<td>MMP-2, -9, -13, TIMP-1, -2</td>
<td>Patients with LV hypertrophy and HF demonstrated specific reductions in MMPs and increased TIMP-1</td>
<td>67</td>
</tr>
<tr>
<td>28</td>
<td>MMP-1</td>
<td>MMP-1 levels reduced in HFpEF</td>
<td>68</td>
</tr>
<tr>
<td>120</td>
<td>TIMP-1</td>
<td>Elevated TIMP-1 associated with reduced early LV filling velocity</td>
<td>69</td>
</tr>
<tr>
<td><strong>Cytokines/chemokines/matrikines</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>118</td>
<td>Cardiotrophin</td>
<td>Increased in patients with robust hypertrophy</td>
<td>70</td>
</tr>
<tr>
<td>52</td>
<td>CTGF, BNP, TGF, MMP-2, TIMP-2</td>
<td>CTGF correlated with TGF, MMP-2, and TIMP-2 and elevated CTGF associated with poor early LV filling velocity</td>
<td>71</td>
</tr>
<tr>
<td>90</td>
<td>Cardiotrophin</td>
<td>Associated with higher filling pressures such as estimated PCWP</td>
<td>72</td>
</tr>
<tr>
<td>202</td>
<td>CXCL16</td>
<td>Increased in HFpEF compared with referent normal subjects</td>
<td>74</td>
</tr>
</tbody>
</table>

AGE indicates advanced glycation end product; BNP, B-type natriuretic peptide; CTP, C-telopeptide for type I collagen; CTGF, connective tissue growth factor; CTX, carboxy-terminal collagen cross-links; CXCL16, chemokine ligand 16; ECM, extracellular matrix; GDF-15, growth differentiation factor 15; HF, heart failure; HFpEF, HF with preserved ejection fraction; HFrEF, HF with reduced ejection fraction; ICTP, carboxy-terminal telopeptide of collagen I; I LV, left ventricular; MMP, matrix metalloproteinase; NT-proBNP, N-terminal pro-brain natriuretic peptide; OPN, osteopontin; PCWP, pulmonary capillary wedge pressure; PICP, procollagen I C-peptide; PIINP, procollagen III N-terminal propeptide; PIINP, procollagen III N-terminal propeptide; PINP, procollagen type I N-terminal propeptide; PIP, procollagen type I; sRAGE, soluble receptor for AGE; TGF, transforming growth factor; and TIMP, tissue inhibitor of MMPs.

*Not intended to be comprehensive but rather representative of studies using peripheral blood sampling.
obtained, indicating a high predictive value for identifying patients with HFpEF. To more carefully examine the predictive value of this ECM biomarker panel in terms of HFpEF prediction, random subsets of patients from the entire cohort were sampled and the prediction algorithm was tested (cross-validation), which was repeated 5 times (5 subsets). The 5 subsets from this cross-validation are plotted to demonstrate conformity to the original AUC curve, confirming the predictive modeling of this biomarker subset for HFpEF. Adapted with permission from Zile et al.66 Authorization for this adaptation has been obtained both from the owner of the copyright in the original work and from the owner of copyright in the translation or adaptation.

Figure 4. Multivariate modeling has been used to identify potential extracellular matrix (ECM) biomarkers to predict the development and progression of heart failure with preserved ejection fraction (HFpEF). A. In this example, a biomarker panel incorporating specific matrix metalloproteinases (MMPs), tissue inhibitors of MMP (TIMPs), collagen peptides, and N-terminal pro-brain natriuretic peptide (NT-proBNP) demonstrated predictive value in patients with HFpEF. Specifically, using a particular cassette of ECM-related plasma biomarkers, which included MMP-2, MMP-8, TIMP-4, and the propeptide of collagen III (PIINP), as well as N-terminal BNP, an area under the curve (AUC) of >0.70 (P<0.01) was achieved. B. As an extension of this example, a biomarker panel including P-selectin (CD62P), N-terminal proBNP, NT-proBNP, PIINP, and 1-Specificity was used to predict the development and progression of HFpEF.

Therapeutics

HFrEF and Targets for Translational Research

The use of biomaterials that are derived from the ECM or mimic a specific composite of the ECM has been the subject of several early translational studies with a particular focus on post-MI remodeling and the progression to HFrEF.81–86 The predominant approach has been to directly inject these ECM biomaterials into the MI region, whereby interruption of MI wall thinning and changes in local stress-strain patterns have been realized.81,82,84–86 For example, a purified bovine collagen containing predominantly type I collagen increased LV wall thickness and relative function in a post-MI rat model.81 In contrast to decellularized or synthesized ECM, synthetic biomaterials with somewhat longer durability and perhaps less direct biological/immune effects have been the focus of investigation in terms of post-MI remodeling.84–86 For example, the use of alginate-based materials injected into the MI region has been shown to attenuate adverse LV remodeling after MI and to change expression profiles of MMPs within the targeted region.82,85 Indeed, the use of an alginate hydrogel–based biomaterial has been advanced to clinical trials in terms of safety and feasibility (A Randomized, Controlled Study to Evaluate Algisyl-LV as a Method of Left Ventricular Augmentation for Heart Failure trial), whereby targeted myocardial injections will be performed using conventional cardiac surgery techniques.87 In addition, the use of both nondurable and durable ECM biomaterials in conjunction with pharmacological agents/bioactive molecules and with a stem cell–type expanded population of cells has also been examined.83,84,88–91 With the use of a polyacrylamide-based hydrogel containing fibroblast growth factor, targeted injections within the MI region...
in rats improved LV pump function. Initial clinical feasibility studies used injection of an ECM biomaterial seeded with bone marrow cells (Myocardial Assistance by Grafting a New Bioartificial Upgraded Myocardium trial) and demonstrated that relative MI thickness was augmented ≤1 year after injection. However, it remains unclear from these initial clinical studies what effects the injection of the processed ECM biomaterial alone may have imparted in terms of endogenous bone marrow cell homing and viability. This holds particular relevance because it has been demonstrated that hydrogel constructs containing hyaluronic acid can influence the viability and chemotaxis of bone marrow–derived stem cells. Nevertheless, ECM biomaterials alone or as an adjunctive approach to local pharmacological or cell-based therapies in the context of post-MI remodeling hold potential therapeutic relevance in the prevention of HFrEF.

Because a prolonged/abnormal local inflammatory state occurs in the post-MI period, interference strategies targeting the inflammation axis would seem to be a logical therapeutic target. However, the strategies to date in terms of targeting specific cytokine pathways or other mediators of inflammation have been either equivocal or surprisingly associated with adverse consequences. For example, inhibition of the chemokine ligand-2 caused a delay in the wound-healing response in post-MI mice, which can lead to myocardial rupture. Thus, further research on the specific inflammatory target and the timing of the specific interference strategy in the post-MI context and how this will directly alter ECM remodeling is required. Targeting downstream effects of inflammation such as the MMPs has also encountered difficulty in implementation in terms of targeting the specific MMP types that are causative in adverse LV remodeling post-MI. For example, broad-spectrum MMP inhibition demonstrated favorable effects with short-term dosing in large-animal models of MI and HFrEF, but concerns about systemic toxicity of broad-spectrum MMP inhibition and difficulties in dosing strategies yielded equivocal clinical results. In addition, genetic deletion of certain MMP types such as MMP-28 has yielded surprising and potentially deleterious effects in transgenic post-MI studies. Nevertheless, because the role of MMP induction and activation in adverse post-MI remodeling is likely significant, the identification of the specific MMP types and selective MMP inhibitors will remain an important area of research. In terms of the activated fibroblast within the MI, the myofibroblast seems to be a TGF-dependent process;
therefore, strategies that disrupt TGF signaling are likely to alter this fibroblast transdifferentiation process.\textsuperscript{14–18,95–97} For instance, membrane-bound MMPs likely contribute to the activation of TGF within the ECM, and this proteolytic activation cascade may yield a potential target.\textsuperscript{18} In other studies, direct interference with TGF by genetic, neutralizing antibodies or oral pharmacological inhibitors has demonstrated early favorable results in animal models of HFrEF.\textsuperscript{95–98}

**HFpEF and Targets for Translational Research**

Although the previous section outlined some potential therapeutic research areas for HFrEF, there are certainly common target areas for HFpEF as well. Because inappropriate ECM accumulation and structure are a milestone in the progression to HFpEF, strategies that target fibroblast form and function such as transdifferentiation to myofibroblasts and TGF interference strategies would be of significance. In addition, directly altering the structure of the ECM with HFpEF will, in turn, alter LV myocardial stiffness properties and LV diastolic function. An example of this strategy is altering posttranslational processing of fibrillar collagen, collagen cross-linking.\textsuperscript{99,100} In both nonhuman primates and initial clinical studies, the glucose cross-link breaker ALT-711 reduced myocardial stiffness associated with LV hypertropy.\textsuperscript{99,100} Interestingly, these beneficial effects of this cross-link breaker were not observed in patients with HFrEF.\textsuperscript{100} These observations underscore the difference in the underlying pathophysiology of these 2 disease processes and the prospective importance of abnormal ECM, particularly fibrillar collagen with HFpEF. In a comparative study of myocardial biopsies, structural differences in fibrillar collagen content and cross-linking were demonstrated in patients with either HFrEF or HFpEF.\textsuperscript{101,102} Although myocardial fibrillar collagen was historically considered to have a relatively slow turnover, collagen turnover can be quite rapid, and this process may be inducible.\textsuperscript{14,103,104} Indeed, the changes in fibrillar collagen peptide fragments, indicative of posttranslational processing, can be quite dynamic in patients with HFpEF and can be affected by pharmacological strategies such as aldosterone receptor antagonists.\textsuperscript{104} Therefore, interruption of profibrotic signaling pathways in the context of developing HFpEF such as inhibition of TGF holds therapeutic potential.\textsuperscript{105,106} In addition to targeting collagen expression by inhibition of profibrotic signaling pathways, interruption of posttranslational processing may be a relevant target for HFpEF. Some examples of matricellular proteins that likely play regulatory roles in ECM structure and function include secreted protein acidic and rich in cysteine, thrombospondin, osteopontin, and peristin.\textsuperscript{16,18,23,95,107–109}

One of the challenges in developing therapeutic strategies for HFpEF is the paucity of biological readouts that may reflect pharmacological efficacy. For example, in patients with a history of hypertension and at risk for developing severe HFpEF, up titration of antihypertensive medications to meet blood pressure goals is a standard of care. Therefore, the use of additional pharmacological agents in patients with the development and progression of HFpEF has been based largely on experience in HFrEF trials, arguably a much different disease process. In other words, the study designs of past HFpEF clinical trials have used empirical dosing strategies, which ultimately have yielded neutral results.\textsuperscript{2,3,5,7,10,30,31} Thus, a rate-limiting step in previous HFpEF clinical trials is that dosing protocols were established through the use of an evidence-based drug titration established on biologically relevant response variables. One future translational and clinical research direction would be to examine the potential pharmacodynamics between ECM biomarkers such as those shown in Table 2 that are likely to be predictive and reflective of the HFpEF progression to titration of pharmacological agents. An example of this concept was recently described in a clinical trial in which patients were treated in response to a change in N-terminal BNP levels from baseline.\textsuperscript{106,111} With this single biomarker used as the therapeutic target and end point, it was established that up titration of standard-of-care medications could effectively reduce this biomarker within ≈3 months and was associated with an improvement in clinical outcomes such as HF hospitalizations. Ongoing studies called Heart Failure (HF) Assessment With B-type Natriuretic Peptide (BNP) In The Home will further test the application of frequent (perhaps daily) assessment of biomarker profiles in tailoring therapy for patients with HFpEF. Thus, future studies that use a specific panel of biomarkers, particularly those related to ECM remodeling, may be reflective of HFpEF progression and severity, which may provide a means of developing a rational dosing strategy and an early readout of efficacy for new therapeutics.

**Summary**

Although basic research efforts have provided unique insights into fundamental processes that govern ECM growth and function, the translation of these findings to improved diagnostics and therapeutics for HF has not been as forthcoming. Several steps that may yield new clinical tools for HF that target the ECM include the need to recognize and differentiate HF phenotypes, as well as the use of biomarkers and multimodality imaging in early translational research studies. Basic and translational research in which a model system is identified simply as HF may be inadequate.\textsuperscript{112,113} Rather, recognizing the classification scheme of HFrEF and HFpEF and incorporating model systems and physiological measurements relevant to these different HF phenotypes, with particular attention to qualitative measures of the ECM biology, would be warranted. A recently performed large population-based study clearly established that HFrEF and HFpEF are 2 distinct HF entities with different natural histories and therefore will likely require different diagnostic and therapeutic approaches.\textsuperscript{114} Thus, coupling new insights into the regulatory pathways and mechanisms of adverse ECM remodeling will likely yield new diagnostic and therapeutic approaches specifically tailored to these forms of HF.

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Spinal and Zile

Myocardial Matrix in Heart Failure Recognition


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91. Purcell BP, Elser JA, Mu A, Margulies KB, Burdick JA. Synergistic effects of SDF-1α chemokine and hyaluronic acid release from degradable hydrogels on directing bone marrow derived cell homing to the myocardium. *Biomaterials*. 2012;33:7849–7857.


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