Chapter 4

Potential clinical application of circulating miRNAs in heart failure

Clinical Use Of Novel Biomarkers In Heart Failure: Towards Personalized Medicine

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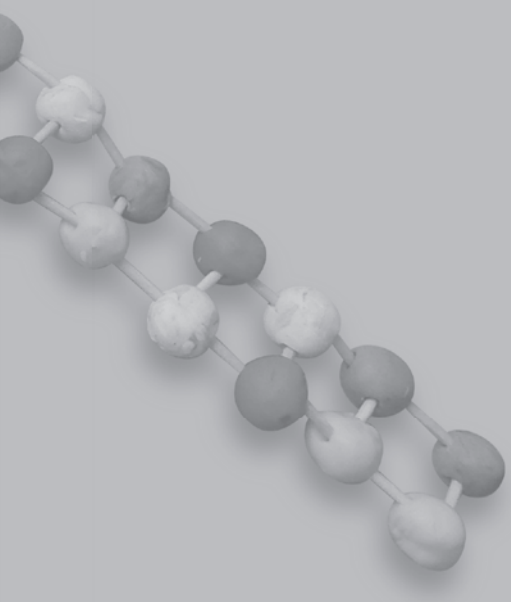

Daniela Schmitter
Gadi Cotter
Adriaan A. Voors

ABSTRACT

Biomarkers play an important role in heart failure. They provide us information about the mechanisms involved in specific types of heart failure and can identify patients at higher risk. Although the majority of biomarker studies in heart failure focus on their prognostic value, the clinical applicability of prognostication in heart failure needs to be established. However, biomarkers can be used for many other purposes. For example, they can help us with the diagnosis of heart failure, and they can be used to select our therapy, leading to personalized tailored therapy. Finally, when biomarkers are causally involved in the disease process, they can even become targets for therapy. The present paper reviews the established and potential value of the novel heart failure biomarkers, mid-regional atrial natriuretic peptide, soluble ST2, growth differentiation factor 15, galectin-3, renal tubular damage markers, and microRNAs. Their potential clinical value will be discussed and compared with the reference markers, the natriuretic peptides.

KEY WORDS

Heart failure, Biomarkers, Personalized medicine, Soluble ST2, Galectin-3, MicroRNAs

INTRODUCTION

In patients with heart failure, biomarkers play an important role, and they are used for the diagnosis and clinical follow-up of heart failure patients. Their importance is reflected by the increasing numbers of scientific papers on biomarkers in heart failure over the last 20 years (Fig. 1). The majority of these papers report on the prognostic value of the biomarker. These studies were often performed on existing databases, using a retrospective analysis. Unfortunately, many studies lack a comprehensive statistical approach and are only based on Cox proportional hazard models. Preferably, the additive value of biomarkers should be established on top of existing prognostic markers, such as age, blood pressure, NYHA functional class, a previous heart failure hospitalization, and heart rate. The statistics should also include comparison of area under the curves of receiver operating characteristics curves, integrated discrimination improvement, and net reclassification index. In the great majority of the papers presented, an external validation in a second cohort is lacking. This could lead to chance findings, related to the specific subset of patients that were studied.

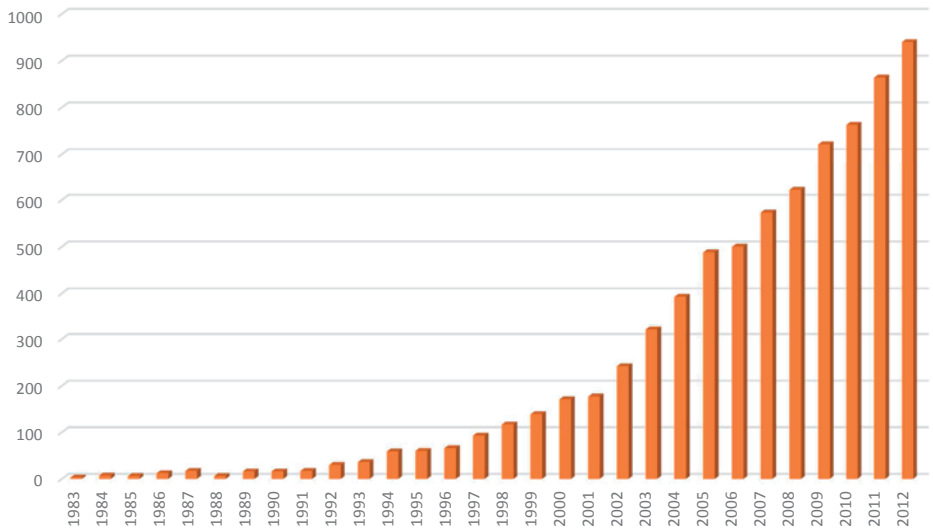

Figure 1. Number of publications on biomarkers in heart failure. Search from PubMed on [biomarkers] and [heart failure].

Nevertheless, some studies are very well performed, and provide adequate statistics, and are validated in several patient populations. However, once the prognostic value of a biomarker on top of existing prognosticators has been established, what does this mean? In other words, what is the clinical use of a good prognostic biomarker, and does the knowledge of a high risk patient change our clinical practice? In general, decision

making of clinicians will change when use of the biomarker leads to a better clinical outcome of the patients. Therefore, prospective clinical trial should be performed, based on a biomarker approach to guide therapy in heart failure. Such trials have been performed with the best-studied biomarkers in heart failure, the natriuretic peptides, brain natriuretic peptide (BNP) and N-terminal pro-hormone of brain natriuretic peptide (NT-proBNP) [1]. However, results were conflicting, with the largest trials not leading to better patient outcome, despite intensification of therapy [1]. Therefore, the current Heart Failure Guidelines of the European Society of Cardiology state that: "High natriuretic peptide concentrations are associated with a poor prognosis, and a fall in peptide levels correlates with a better prognosis. However, several randomized clinical trials that evaluated natriuretic peptide-guided treatment have given conflicting results. It is uncertain whether outcome is better using this approach than by simply optimizing treatment." [2] This shows that there is a major gap between establishing the prognostic effects of a biomarker toward showing beneficial clinical effects of a biomarker-guided approach.

Fortunately, this does not mean that biomarkers cannot be of use to clinicians. There are many other potential clinical applications in which biomarkers can be useful in heart failure patients, finally leading to a better clinical outcome (Table 1). First, biomarkers can provide us with a better insight in the pathogenesis of heart failure. For example, biomarker research has taught us that heart failure is strongly related to an activation of the renin angiotensin aldosterone system (RAAS) and the sympathetic nerve system. These insights have resulted in the most important therapies in heart failure, RAAS-blockers and beta-blockers. Second, biomarkers might help us in the diagnosis of heart failure. The best-known examples are the natriuretic peptides. Several studies have indicated that the use of natriuretic peptides at the emergency department results in a better and quicker diagnosis, leading to reduced length of hospital stay and hospital costs [3, 4]. Third, biomarkers might help to better target our therapeutic approach [5]. For example, studies have indicated that galectin-3 predicts the response to a statin in patients with chronic heart failure [6]. These and other studies might result in a personalized therapeutic approach. Finally, biomarkers themselves might become a target of therapy. For example, human recombinant forms of natriuretic peptides, such as nesiritide, have been studied as a treatment in acute decompensated heart failure [7]. Therefore, biomarkers can be a useful tool for many reasons other than to improve prognostic accuracy. Here, we will review potential clinical applications of novel biomarkers in patients with heart failure.

Table 1. Potential roles for circulating miRNAs as biomarkers in heart failure

Disease	Study Design	miRNA Biomarkers	Source	Diagnostic potential	Prognostic potential	Response to therapy	Comments	Reference
HF	30 HF 20 non-HF with dyspnea 39 healthy subjects	miR-423-5p↑ Others: miR18b*, miR-129-5p, miR-1254,HS_202.1, miR-622, miR-654-3p	Plasma	miR-423-5p: HF vs healthy controls AUC = 0.91 HF vs non-HF with dyspnea AUC = 0.83			miR-423-5p relation to disease severity: LVEF, NYHA class, NTproBNP	[65]
AHF	33 AHF 34 healthy controls	miR-499↑, miR-122↑	Plasma				In patients with acute heart failure, only miR-499 was significantly elevated (2-fold).	[67]
HF	10 HF 17 asymptomatic controls	miR-126 ↓ Not changed: miR-122, miR-499	Plasma				miR-126 relation to disease severity: negatively correlated with age, logBNP and NYHA class	[66]
HF	9 MI, 5 unstable AP, 15 HF; 10 healthy subjects	miR-499 not changed	Plasma				miR-499 increased in AMI but was below detection limit for all individuals in the other patient groups including HF	[76]
HF	30 CHF 30 healthy controls	miR-423-5p, miR-320a, miR-22 miR-92b = miRNA score	Plasma	miR-423-5p: HF vs healthy controls AUC = 0.88 miRNA score: HF vs healthy controls AUC = 0.90			miR-423-5p relation to disease severity: BNP but no correlation to LVEF and NYHA class; miRNA score correlation to functional parameters: elevated BNP serum levels, a wide QRS, dilatation of the left ventricle and atrium	[64]
HF	41 right ventricular HF 10 healthy controls	miR-423-5p unchanged	Plasma				In patients with right ventricular HF and reduced EF miR-423-5p levels are not elevated	[77]

Table 1. Potential roles for circulating miRNAs as biomarkers in heart failure (continued)

Disease	Study Design	miRNA Biomarkers	Source	Diagnostic potential	Prognostic potential	Response to therapy	Comments	Reference
HF	15 ischemic cardiomyopathy ICM 19 nonischemic dilated cardiomyopathy NIDCM 19 healthy controls	miR-107, miR-139, miR-142-5p, miR-142-3p, miR-19b, miR-125b, miR-497	PBMC	miR-107, miR-142– 5p, and miR-139 ↓ in both classes of HF; miR-125b, miR-497 ↓ in ICM only; miR-142, miR-29b ↑in NIDCM only			miRNAs specifically regulated in the PBMCs of NIDCM and ICM patients with a potential diagnosic and/or prognostic use in CHF	[78]
HF	8 diastolic function and preserved systolic function (DD) 10 stable compensated dilated cardiomyopathy (DCM—systolic plus diastolic dysfunction) 13 decompensated congestive heart failure (DCM–CHF—systolic plus diastolic dysfunction) 8 healthy controls	miR-454, miR-500, miR-1246, miR-142-3p, miR-124-5p	Buffy coat	miR-454, miR-500 ↓in DD; miR-1246 ↑ in DD; miR-142-3p ↓in DCM and DCM–CHF but not in DD; miR-124-5p ↑↑in DCM but not in DD and DCM–CHF			miRNA correlation to functional parameters: BNP, Ea (velocity of early myocardial relaxation), E/Ea (representing) left atrial pressure; LVEF in DCM group	[79]
HF	Rat model of HF	miR-499-5p, miR-423-5p	Cardiac tissue and plasma			HF hypertension induced in Dahl saltsensitive rats. Treatment with antimiR-208a improved cardiac function and survival. Anti-miR treatment blunted increase of circulating miR-499-5p and miR-423-5p levels	miR-208 as therapeutic target for the modulation of cardiac function and remodeling during heart disease progression	[73]

MR-proANP

(Patho)physiology

Natriuretic peptides have become well-established markers both for the diagnosis and for the prognosis of patients with acute and chronic heart failure. The two best-studied natriuretic peptides are brain natriuretic peptide (BNP) and the N-terminal pro-BNP (NT-proBNP). Atrial natriuretic peptide is derived from the cleavage of its precursor pro-atrial natriuretic peptide (proANP), which is significantly more stable in the circulation than the mature peptide. Therefore, proANP is suggested to be a more reliable biomarker. However, the N- and C-terminal regions of propeptides still can undergo enzymatic degradation, and therefore, a new sandwich immunoassay that recognizes a mid-regional sequence of proANP (MR-proANP) is even be more stable. This assay has been extensively tested in patients with heart failure.

Prognosis

Several studies showed similar prognostic value of MR-proANP compared with BNP and NT-proBNP in patients with chronic and acute heart failure [8–11]. A strategy of serial monitoring of MR-proANP further increased its prognostic accuracy [12].

Diagnosis

The diagnostic value of MR-proANP has been thoroughly studied in patients with acute heart failure and compared with BNP and NT-proBNP. The BACH (Biomarkers in Acute Heart Failure) trial was a prospective study of 1,641 patients presenting to the emergency department with dyspnea [13]. In this trial, MR-proANP appeared to be as useful as BNP for the diagnosis of acute heart failure in dyspneic patients. Similar findings from several other studies confirmed these findings [14, 15]. Based on this, MR-proANP is now the second biomarker that is recommended in the 2012 Heart Failure Guidelines of the European Society of Cardiology as an alternative for the diagnosis of heart failure in patients with acute dyspnea presenting at the emergency department [2].

Unlike BNP and NT-proBNP, no studies have been performed to establish the effects of MR-proANP-guided therapy in heart failure patients.

ST2

(Patho)physiology

ST2 is a member of the interleukin 1 receptor family. The ST2 protein has two isoforms: a soluble form (referred to as soluble ST2 or sST2) and a membrane-bound receptor form (referred to as the ST2 receptor or ST2L). The ligand for ST2 is the cytokine interleukin-33 (IL-33). Some studies indicated the cardioprotective effects of IL-33 [16]. ST2L mediates

the effects of IL-33, whereas sST2 limits the activity of IL-33. It has been speculated that increased levels of the active form of IL-33 in CHF patients may have protective effects in the progression of heart failure and may reduce oxidative stress [17]. However, so far ST2 and IL-33 have not significantly improved our understanding of the pathogenesis of heart failure.

Prognosis

A large amount of studies have shown that elevated levels of ST2 are related to a higher risk of mortality, both in acute decompensated heart failure and in chronic heart failure patients [18–23]. These studies also showed some correlation between ST2 and the natriuretic peptides, although ST2 had an additive prognostic effect on top of the natriuretic peptides [24]. In addition, an increase in ST2 over time in patients hospitalized for acute heart failure was also related to a poorer outcome [25]. Based on these findings, the ST2 assay has been recently approved by the United States Food and Drug Administration for use as an aid in assessing the prognosis of patients diagnosed with chronic heart failure.

Diagnosis

Conflicting results have been published on the diagnostic value of ST2 to identify patients with heart failure. In a study from Aldous et al., 995 patients attending the Emergency Department with chest pain were prospectively recruited [26]. The diagnostic value of ST2 to detect heart failure was similar to BNP, but BNP failed to identify 4 of 35 individuals with a primary outcome, and 2 of them were identified by ST2. So, the combination of both might provide an even more accurate diagnosis of heart failure. However, in a previous study by Januzzi et al. in patients presenting to the emergency department with dyspnea, the diagnostic value of NT-proBNP was superior to ST2 for diagnosis [18]. This was confirmed in another study in 251 consecutive patients presenting to the emergency department with dyspnea, where the diagnostic value of ST2 was inferior compared with BNP and MR-proANP [15]. Therefore, ST2 seems to be of limited value in the diagnosis of heart failure.

So far, no studies are published on the potential roles of ST2 in guiding therapy or as a direct target for therapy.

GDF-15

(Patho)physiology

Growth differentiation factor 15 (GDF15) is a protein belonging to the transforming growth factor beta superfamily that has a role in regulating inflammatory and apoptotic pathways in injured tissues. Although GDF15 is weakly expressed in the heart and

other tissues under physiological conditions, its expression may increase significantly in response to inflammation and tissue injury. In a study comparing patients with heart failure with preserved left ventricular ejection fraction (HFpEF) and reduced ejection fraction (HFrEF), GDF15 was expressed more pronounced in HFpEF patients [27]. This might suggest that HFpEF is more related to systemic inflammation, while in patients with HFrEF, biomarkers of myocardial injury (hsTnT) and increased wall stress (NT-proBNP) were elevated more pronounced.

Prognosis

A few studies have demonstrated that elevated levels of GDF15 were related to a higher mortality in patients with chronic heart failure [28, 29]. In addition, GDF15 provided independent additional information on top of established prognosticators and other biomarkers, including the natriuretic peptides. Also, increases in GDF-15 over 12 months were independently associated with the risks of future mortality and first morbid event in patients with chronic heart failure [30].

Diagnosis

Interestingly, the diagnostic properties of GDF15 have been better studied for HFpEF patients than for HFrEF patients. In a study with morbidly obese individuals, GDF15 levels seemed to better correlate with diastolic dysfunction than NT-proBNP levels, and GDF15 significantly improved reclassification for the diagnosis of heart failure and added incremental value to NT-proBNP [31]. In another study, the diagnostic properties of GDF15 for detecting HFpEF tended to be superior to those of NT-proBNP, and a combination significantly improved diagnostic accuracy [32].

Similar to ST2, no studies are published on the potential roles of GDF15 in guiding therapy or as a direct target for therapy.

Galectin-3

(Patho)physiology

Galectins are a family of soluble b-galactoside-binding lectins that play many important regulatory roles in inflammation, immunity, and cancer. Recently, a role for galectin-3 in the pathophysiology of heart failure has been suggested [33]. Several studies have demonstrated the association between levels of galectin-3 and macrophage migration, fibroblast proliferation, the development of fibrosis, and left ventricular hypertrophy.

Prognosis

Multiple studies have shown prognostic effects of galectin-3 in patients with both acute [34–36] and chronic heart failure [37–42]. The value was incremental to established

prognosticators and other biomarkers, including NT-proBNP. Only one study reported that the predictive value of galectin-3 disappeared after adjustment for NT-proBNP [43].

Diagnosis

The diagnostic value of galectin-3 for heart failure has been less well established. In 599 patients presenting with dyspnea at the emergency department, 209 (35%) patients had acute heart failure [34]. In this cohort, NT-proBNP was superior to galectin-3 for the diagnosis of acute HF, although galectin-3 levels were significantly higher in subjects with heart failure compared with those without.

Guiding therapy

In 1492 patients with ischemic chronic heart failure, there was a significant interaction between the level of galectin-3 and the effects of statin therapy on the primary endpoint of cardiovascular death, myocardial infarction, or stroke [6]. Similarly, since galectin-3 is a fibrosis marker, it has been suggested that patients with heart failure and raised levels of galectin-3 might benefit more from aldosterone antagonist therapy than patients with lower levels [44]. Although this hypothesis needs to be confirmed in a prospective trial, experimental data already indicate that galectin-3 mediates aldosterone-induced vascular fibrosis [45]. Therefore, galectin-3 might be useful to indicate specific patients that benefit from heart failure therapy, which could be a first step toward personalized medicine.

Target for therapy

There is accumulating evidence that galectin-3 is causally involved in the development and progression of heart failure. Disruption of the galectin-3 gene blocked myofibroblast activation and procollagen expression in vitro and in vivo, and addition of exogenous recombinant galectin-3 in vitro reversed this abnormality [46]. A potential causative role for galectin-3 in the development of heart failure is further supported by data from 3,353 participants in the Framingham Offspring Cohort Framingham study indicating that higher levels of galectin-3 in the general population are related to new onset heart failure [47]. It has therefore been suggested that specific anti-galectin therapy may halt the development of heart failure [44], and anti-galectin studies in heart failure are currently ongoing.

Renal tubular markers

(Patho)physiology

Renal dysfunction is common in patients with heart failure and is associated with high morbidity and mortality [45–50]. In general, renal dysfunction is defined as an impaired

glomerular filtration rate, reflected by an increase in serum creatinine or cystatin-C. However, renal failure is not only limited to impaired filtration but also includes glomerular hypertension and tubulointerstitial hypoxia, leading to loss of glomerular integrity and tubular damage. Experimental studies have shown that impaired renal perfusion in heart failure predisposes to hypoxic outer medullary injury as well, which might predispose to tubulointerstitial hypoxic damage [51]. Although multiple renal tubular markers are currently under investigation, three renal tubular markers have been studied in heart failure: Neutrophil gelatinase-associated lipocalin (NGAL), N-acetyl-beta-d-glucosaminidase (NAG), and Kidney injury molecule-1 (KIM-1) [52]. While NGAL can be measured both in urine and in plasma, KIM-1 and NAG can only be measured in urine. Due to the important role of renal function in heart failure, and its distinctive characteristics, assessments of both plasma and urinary concentrations of markers for tubular injury can be of clinical use in patients with heart failure.

Prognosis

The predictive value for clinical outcome in heart failure for NAG, NGAL, and KIM-1 has been well established in different cohorts of patients with acute decompensated heart failure [53–57]. Interestingly, their predictive value remained present after adjustment for creatinine (or estimated glomerular filtration rate—eGFR) and albuminuria, further indicating the differential role of tubular markers compared with glomerular markers.

Diagnosis

In patients with normal eGFR (>60 ml/min), tubular markers might still indicate renal damage in patients with heart failure. However, they are not suitable as a marker for the diagnosis of heart failure.

Guiding therapy

Tubular markers are sensitive markers to small hemodynamic changes. In stable chronic heart failure patients, small volume changes were not associated with changes in creatinine, but they were clearly associated with changes in renal tubular markers [58]. Therefore, they might support clinical decision making in unstable heart failure patients. In addition, elevated serum NGAL levels, measured at the time of hospital admission for acute decompensated heart failure, can predict the development of worsening renal function [59]. This might be related to a much faster release of tubular markers in the circulation compared with creatinine, which is known to be a slow marker that is only increased after 24 h of renal injury [60]. Early detection of patients at risk for worsening renal function in patients admitted for acute decompensated heart failure might result in the prevention of worsening of renal function, potentially leading to shorter length of hospital stay and improved clinical outcome. However, these potential applications

of renal tubular markers need to be established in well-designed prospective renal-marker-guided therapy studies.

Target for therapy

Although it has been suggested that renal failure is causally related to heart failure, so far no study convincingly showed that directly improving renal function will lead to better outcomes in patients with heart failure [61].

MicroRNAs

(Patho)physiology

MicroRNAs are a class of noncoding small RNAs that regulate gene expression at the posttranscriptional level. Therefore, microRNAs can regulate protein expression as shown in Fig. 2. Many of these microRNAs are present in the heart and dynamically regulate the response to acute cardiac stress and in some cases during long-term compensatory response of the heart to a chronic injury or hemodynamic overload [62, 63]. Interestingly, the expression pattern of microRNAs changes significantly when the severity of symptoms of heart failure increases (Table 1). Two independent studies with chronic heart failure patients consistently showed up-regulation of miR-423-5p compared to healthy control cohorts and patients with dyspnea but no heart failure [64, 65]. In addition, some studies related the expression of certain microRNAs to markers of severity of

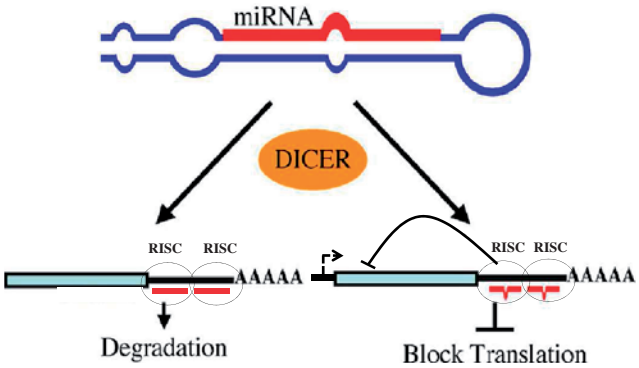

Figure 2. MicroRNAs mechanism of action. MicroRNAs are produced from either genes or from introns. After transcription, nuclear processing and export, the pre-miRNA hairpin (as shown above) is cleaved by the RNase III enzyme Dicer in the cytoplasm. The imperfect miRNA:miRNA* duplex is about 22 nucleotides in length. Although either strand of the duplex may potentially act as a functional miRNA, only one strand is usually incorporated into the RNA-induced silencing complex (RISC). The miRNA-RISC complex binds to the target mRNA in a sequence-specific manner, inducing cleavage and degradation or preventing the binding of ribosomes. This process is known as gene silencing and leads to depletion of protein levels.

heart failure, such as NYHA functional class and natriuretic peptides [64, 66–79]. Hence, microRNAs may play an important role in the pathogenesis of heart failure.

Although microRNAs are primarily present and studied in tissues, such as the heart, circulating microRNAs are now emerging as blood-based biomarkers also for heart failure. MicroRNAs offer many attractive features as biomarkers. They are stable in the circulation, their sequences are evolutionarily conserved, their expression is often tissue or pathology specific, and their detection is based on sequence-specific amplification, features that are helpful in the development of sensitive and specific assays.

Prognosis

To our knowledge, there is currently no study published that has related certain microRNAs to the prognosis in acute and chronic heart failure patients with regard to hard clinical endpoints such as heart failure hospitalization and death. In a few studies, the correlation between microRNAs and disease severity and progression of the disease has been assessed, as shown in Table 1.

Diagnosis

In a study published in 2010 by Tijsen et al., miR-423-5p distinguished patients with heart failure from healthy controls with an AUC of 0.91 and from patients with dyspnea but without heart failure with an AUC of 0.83 [65]. Goren et al. confirmed that circulating miR-423-5p levels were elevated in 30 patients with chronic heart failure compared with 30 age-, sex-, and ethnically matched healthy controls [64]. From 186 microRNAs that they studied, 26 showed significantly different levels in patients with HF, of which miR-423-5p showed the strongest increase. In this study, miR-423-5p was able to distinguish patients with heart failure from healthy controls with an AUC of 0.88 [64]. However, evidence is lacking that increased plasma levels of miR-423-5p originate from the failing heart or from other organs through different release mechanisms. When Goren et al. used a panel of four specific microRNAs (miR-423-5p, miR-320a, miR-22, and miR-92b), diagnostic accuracy was further improved with an AUC of 0.90 [64]. Several other microRNAs were found to be specifically enriched in chronic heart failure patients [67] but more conclusive studies in larger populations are needed to select the microRNAs with the highest potential to become diagnostic biomarkers in heart failure.

Guiding therapy

In heart failure patients who received a left ventricular assist device, tissue microRNAs expression profile was mostly normalized in accordance with the response to this therapy [68]. However, the need for an invasive procedure to obtain myocardium samples makes the clinical application very limited [69]. Also, this study only supports the hypothesis that microRNAs are related to markers of the severity of heart failure, but further studies

need to be performed to establish the potential value of microRNAs to guide therapy in heart failure.

Target for therapy

Interestingly, microRNAs might become a target for therapy. Aberrant microRNA expression can be normalized by two different ways: by the use of antagomirs and miR-mimics [70, 71]. Antagomirs can silence microRNAs that are over-expressed in heart failure, and miR-mimics can replace those that have a deficit in expression in heart failure. Their mode of action is described in Fig. 3. In an experimental mice model of cardiac hypertrophy, microRNAs were over-expressed in cardiac fibroblasts [72]. The use of an antagomir designed to functionally inhibit miR-21 significantly reduced cardiac hypertrophy and fibrosis and improved cardiac function [72]. Similarly, systemic delivery of antimiR-208a in a Dahl salt-sensitive rat heart failure model resulted in improved cardiac function and survival [73]. Therefore, antagomirs might become useful therapies to prevent or reverse cardiac hypertrophy and improve cardiac function. However, these therapies are based on only silencing single microRNAs. Since multiple microRNAs are likely to be

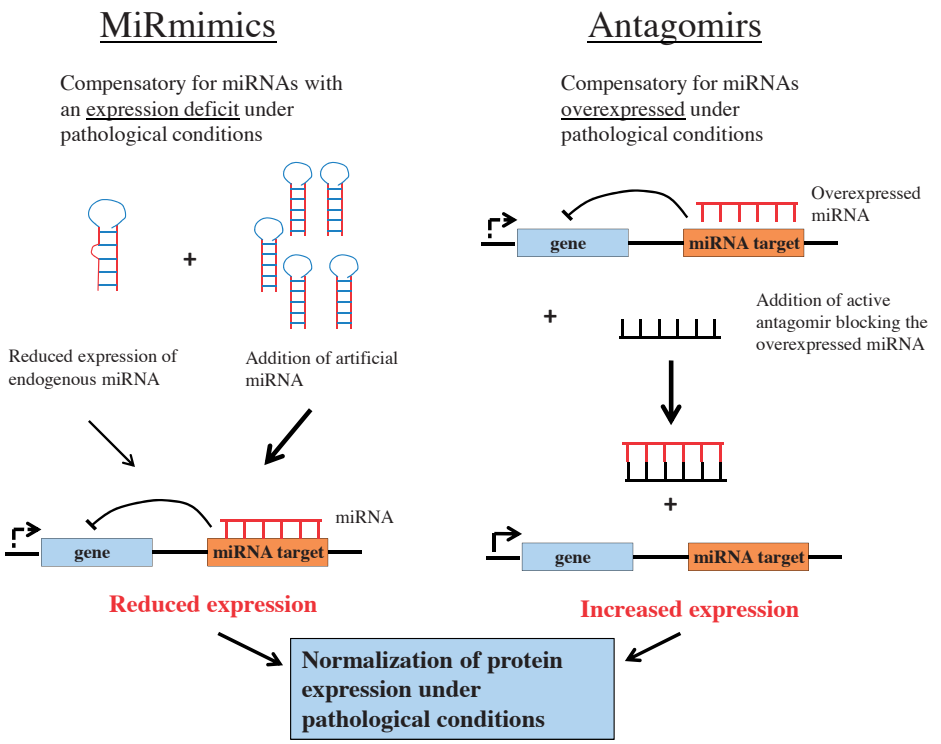

Figure 3. Schematic overview of two treatment strategies normalizing miRNA and hence protein expression under pathological conditions.

involved in the development and progression of heart failure, several microRNAs must be silenced to obtain an effective therapy. A second way to use microRNAs as a target for therapy is the use of miR-mimics [73]. In an experimental rat model of pressure overload, miR-mimics resulted in a normalization of cardiac dilation and a significant reduction of cardiac hypertrophy, cardiomyocyte diameter, and cardiac fibrosis [74].

Currently, the potential of microRNAs as therapeutic targets is also under investigation in clinical trials. The most advanced drug discovery program focused on developing microRNA-based therapeutic targets is based on miRNA-122, expressed in the liver, using the locked nucleic acid (LNA)—modified antisense oligonucleotide miravirsen [75]. Miravirsen is currently studied in phase 2 clinical trials for the treatment of hepatitis C virus (HCV) infection. Data from a phase 2a study indicate that a four-week miravirsen monotherapy provides long-lasting suppression of viremia, has a high barrier to viral resistance, and is well tolerated in patients with chronic HCV infection [75]. To our knowledge, so far no clinical heart failure studies with antimiRs or miR-mimics are ongoing.

DISCUSSION

Table 2. Evidence based indications for the use of novel biomarkers in heart failure

	Prognosis	Diagnosis	Pathophysiological Insight	Guide for Therapy	Target for Therapy	Changes clinical practice
BNP/NT-proBNP	+++	+++	++	+/-	-	++
MR-proANP	++	+++	+	U	U	+
ST-2	+++	+	-	-	-	-
GDF-15	++	++	+	U	U	-
Galectin-3	+++	-	++	+	I	-
Tubular markers	++	-	+	I	-	-
Micro-RNAs	-	+	+	-	I	-

+ = positive; - = negative; U = unknown; I = currently under investigation

Over the last 20 years, there is a steady increase in the number of publication on biomarkers in heart failure (Fig. 1). The large majority of these papers are related to the prognostic value of these biomarkers. However, a prognostic marker will generally not improve clinical decision making. Tremendous efforts of (NT-pro)-BNP-guided therapy trials in heart failure have not resulted in a recommendation in the heart failure guidelines for its clinical use. However, besides the prognostic value, many other interesting potential applications of biomarkers in heart failure exist (Table 2).

First, there are now three well-established markers that can support or reject the diagnosis of heart failure: BNP, NT-proBNP, and MR-proANP. The latter is only recommended

in patients with acute heart failure. The cutoff points that are used for these markers in the recent ESC Heart Failure Guidelines are based on the optimal cutoff point provided by ROC analysis [2]. A consistent finding is that at the optimal cutoff point, sensitivity is always higher than specificity. This implicates that these markers are better to rule out the diagnosis of heart failure, than to prove or establish its diagnosis. Therefore, there is still room for the improvement for more specific biomarkers for heart failure.

Second, biomarkers might help us to guide therapy. For example, some therapeutic approaches may be more effective in patients with specific biomarker profiles. So far, such a strategy has been studied with natriuretic peptides. These (NT-pro)-BNP-guided studies assumed that patients with elevated levels would benefit more from intensified treatment than patients with lower levels, which yielded conflicting results. Some studies showed a beneficial effect, while others were neutral, or only positive in patients aged <75 years of age. Taken together, two meta-analyses showed a potential beneficial effect of natriuretic peptide-guided therapy, although this was not yet translated into a clear guideline recommendation. Therefore, one could consider the current natriuretic peptide studies as pilot studies and that the meta-analyses support more definitive prospective large-scale outcome trials of the strategy which are underway. However, the conflicting results might also have been caused by a false hypothesis. There is some evidence that therapy might even be better in patients with lower levels, since patients with the highest levels (and the highest risk) might be "beyond repair" [80].

This approach of "therapy guidance" should be clearly distinguished from biomarkers used for therapy selection. Using biomarkers for the selection of therapy is based on the pathophysiological background of the biomarker. For example, vasopressin antagonists might be particularly beneficial in patients with acute decompensated heart failure and low serum sodium levels [81]. Another example is that RAAS-inhibitors are particularly beneficial in patients with high plasma renin activity, as has been shown in patients with hypertension [82]. A biomarker-guided approach might therefore lead to personalized medicine, with obvious advantages on clinical outcome, reduction of side effects, and healthcare-related costs.

Third, an obvious advantage of biomarker research is our increased understanding of the pathogenesis of heart failure. The best-proven drugs in heart failure, RAAS-inhibitors and beta-blockers, have been found after establishing that these biomarkers are strongly elevated in heart failure, related to markers of the severity of heart failure and clinical outcome. Therefore, the ideal outcome of biomarker research is that it might lead to direct targets for therapy.

Fourth, ideally biomarkers itself might become a target for therapy. One example is galectin-3, and first clinical studies with anti-galectin therapy are ongoing. Other interesting examples are microRNAs. MicroRNAs will further improve our understanding on proteins that are involved in the pathogenesis of heart failure. This might lead to clinical

heart failure studies with antagomirs or miR-mimics that might target processes that are typical for specific types of heart failure, since it is well known that heart failure is not a disease but a manifestation of diverse cardiac and non-cardiac abnormalities. However, before microRNAs become reality as established biomarkers and therapeutic targets, many studies are still needed in order to overcome important obstacles. First, so far studies have been small and were not confirmed in validation cohorts. Also, their value should be compared with established biomarkers. Second, no standardized method for the detection and quantification of circulating microRNAs in plasma and serum samples has been established. Third, the choice of normalization is critical for evaluating circulating microRNA levels by qRT-PCR strategies. Finally, efforts to better understand the biological processes controlling microRNA release and stability are needed and might be of great interest.

Therefore, biomarker research in heart failure will continue to play an important role. However, the focus should shift more from their prognostic value, toward a better understanding of the pathogenesis of heart failure. It should be emphasized that heart failure is a clinical syndrome, involving the heart and many other organs [83]. Biomarkers might help us to better understand the pathophysiology of heart failure. Therefore, different biomarkers will have different meanings and implications in specific expressions of heart failure. A better understanding might result in specific biomarker-targeted therapeutic approaches and ideally novel targets for therapy, leading us toward personalized therapeutic treatment of patients with acute and chronic heart failure.

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CONFLICTS OF INTEREST

None.

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