REVIEW TOPIC OF THE WEEK

A Test in Context
Critical Evaluation of Natriuretic Peptide Testing in Heart Failure

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CME Objective for This Article: After reading this article, the reader should be able to: 1) Relate the importance of knowing the clinical context when assessing natriuretic factor levels in patients with various stages of heart failure; 2) Discuss the difference between BNP and NT-proBNP levels when assessing heart failure in patients receiving nephrin inhibitors; and 3) Describe the current role of sequential natriuretic level measurements in monitoring therapy of patients with heart failure.

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A Test in Context

Critical Evaluation of Natriuretic Peptide Testing in Heart Failure

ABSTRACT

Circulating natriuretic peptide measurements have been used extensively over the past 15 years to diagnose and monitor patients with heart failure. We are still learning how complex the dynamics of natriuretic peptides can be in the interpretation of test results in individual patients. Although natriuretic peptide measurements are widely used in practice, there are questions regarding why these peptides may not necessarily track with blood volume or invasive hemodynamic measurements in individual patients. Interpretation of natriuretic peptide measurements will depend on many factors, including special patient populations, obesity, renal function, the state of congestion or decongestion, and whether patients are receiving specific therapies. Natriuretic peptide measurements have clearly revolutionized clinical care for patients with heart failure, but further research should provide insights to help use these measurements to individualize patient care beyond the current guidelines. (J Am Coll Cardiol 2016;67:330–7) © 2016 by the American College of Cardiology Foundation.

The first measurements of circulating atrial natriuretic peptide (ANP) in patients occurred in the early 1980s (1). Burnett et al. (1) from the Mayo Clinic measured circulating ANP in human volunteers and patients with various cardiovascular diseases, including heart failure (HF). These measurements built on previous work of de Bold et al. (2), who reported that injecting atrial myocardial extracts intravenously into rats produced a rapid and potent natriuretic response. Burnett et al. (3) had previously reported a decrease in blood pressure and inhibition of the renin-angiotensin-aldosterone system with injection of synthetic ANP into animals, but it was their later seminal clinical research work that introduced us to the concept of a circulating biochemical marker as a means to "diagnose" increased filling pressure in patients with heart disease. Original studies by Troughton et al. (4), Maisel et al. (5), Januzzi et al. (6), and others translated these early observations into clinical practice by demonstrating that natriuretic peptides are extremely useful in the diagnosis of HF, essentially changing the clinical approach to the care of the patient with HF.

The idea of measuring a circulating biomarker to measure the presence and severity of HF was on its way to what has become an interesting journey. Moving forward from the early 1980s to the present time, when natriuretic peptides (primarily B-type natriuretic peptide [BNP] and its inactive fragment, N-terminal pro-B-type natriuretic peptide [NT-proBNP]) are widely used to aid clinical decision making in patients with HF, sets the stage for a critical appraisal of where we are today with these biomarkers. It raises the question of whether we are really better off today with the publication of the seminal Burnett et al. (1) paper in Science than we were in 1986. How should we now use and apply these biomarkers today in the care of our patients with shortness of breath and/or HF?

We now know much more about ANP, BNP, and NT-proBNP than before. At present, biologically inactive NT-proBNP and BNP are the most widely-used diagnostic biomarker tools for the evaluation of patients with dyspnea and/or HF. Careful clinical evaluation/reasoning and interpretation of the biomarker in the correct context will always trump a biomarker test alone in patients with the complex syndrome of HF. Clinical decision making is a craft involving numerous factors, including experience and an understanding of scientific evidence. It must be fine-tuned when dealing with individual patients, and should be tailored to specific clinic settings (such as the emergency department), or to special patient populations (e.g., those with renal dysfunction). The measurement of any biomarker in isolation, without clinical context, will never be acceptable. Nevertheless, natriuretic peptide measurements are readily available and widely used, but still do not always provide clear-cut guidance regarding specific therapies.

WHAT ARE NATRIURETIC PEPTIDES?

The late 1980s and early 1990s produced a huge amount of literature on natriuretic peptides. Marked elevation of cardiac filling pressure, with or without the syndrome of HF, is clearly accompanied by higher
concentrations of circulating natriuretic peptides. BNP is produced as a pre-prohormone, then processed to proBNP, which is cleaved by corin to produce biologically active BNP and inactive NT-proBNP. BNP (but not NT-proBNP) is one of many substrates degraded by neprilysin (a fact that has recently increased in clinical relevance with the results of PARADIGM-HF [Prospective Comparison of Angiotensin Receptor-Nepriylisin Inhibitor with Angiotensin-Converting-Enzyme Inhibitor to Determine Impact on Global Mortality and Morbidity in Heart Failure Trial]; see later discussion of wet and dry natriuretic peptide levels).

With clinical experience, it soon became clear that sodium retention and peripheral edema still occur in patients with HF, despite elevated circulating natriuretic peptide levels. This seemed paradoxical, as high-circulating natriuretic peptide levels were thought to counteract the syndrome of HF. Circulating natriuretic peptide assay levels may include a mix of NT-proBNP, BNP fragments, ANP, and C-type natriuretic peptide. Some of these may be less biologically active than BNP. We knew that ANP was located in the specific cardiac myocyte granules first described by Jamieson and Palade in 1964 (7). These are membrane-bound, polypeptide-hormone-storing granules, 250 to 500 μm in diameter. ANP and BNP are often found coexisting in the same storage granule. It is not clear why some BNP is stored in a processed form, whereas ANP is stored mainly unprocessed. In general, BNP shares a similar biological spectrum of activity with ANP. Under certain conditions where there may be continuous stimulation of natriuretic peptides (as may occur in severe HF), there may be differences in the production of these hormones. Both ANP and BNP receptors activate guanylate cyclase to generate cyclic guanosine monophosphate, which results in vasodilation. The C-receptor, sometimes referred to as the natriuretic peptide clearance receptor, probably signals through G-proteins, rather than cyclic guanosine monophosphate. Natriuretic peptides also interact with adrenal glomerulosa, where they reduce aldosterone production via a change in ion channel activity. Both the synthesis and the release of aldosterone are inhibited by natriuretic peptides. The sympathetic nervous system is also inhibited by natriuretic peptides. ANP reduces renin secretion and arginine vasopressin secretion. Both ANP and BNP inhibit release of endothelin-1 from endothelial cells. Both have antigrowth properties in the vasculature. A recent review described the current use of natriuretic peptides in the syndrome of HF (8).

**CONTROL OF NATRIURETIC PEPTIDE RELEASE**

ANP and BNP are both continuously released from the heart. Atrial muscle stretch is well known to augment ANP and BNP release, although the precise mechanism is unknown. Nevertheless, heightened release of ANP and BNP is a direct consequence of mechanical stretch of atrial muscle (9). It may be that increases in atrial dimension, rather than changes in atrial pressure, stimulate natriuretic peptide release (9). This concept is supported by the observation that in patients with cardiac tamponade, the high pressure, but low volume in the atria is associated with low-normal ANP levels, whereas pericardiocentesis results in elevated ANP (10). ANP is also elevated in patients with ongoing supraventricular tachycardia and atrial fibrillation in the absence of HF.

ANP is released mainly from the atria. ANP messenger ribonucleic acid reaches only modest ventricular levels (7%), relative to atrial levels (11). Unlike ANP, BNP resides primarily in ventricular muscle. BNP messenger ribonucleic acid content is much higher in the ventricle. BNP is cleared more slowly from the circulation than ANP. BNP release is also stimulated by endothelin-1 and phenylephrine. BNP and NT-proBNP are mainly products of the ventricular myocytes. Most hospitals now use NT-proBNP or BNP assays to measure natriuretic hormone levels. Asian and Black patients with HF have higher natriuretic peptide levels on admission compared with White and Hispanic patients (12). Patients with severe HF have markedly increased myocardial tissue concentrations of ANP that parallel the increased circulating levels (13). Obese patients tend to have low circulating natriuretic peptide levels for a given

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**TABLE 1 Current Indications for Natriuretic Peptide Measurements in HF**

<table>
<thead>
<tr>
<th>Indication</th>
<th>ACC/AHA Recommendation</th>
<th>Class-Level of Evidence</th>
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</thead>
<tbody>
<tr>
<td>Diagnosis in patients with dyspnea (acute)</td>
<td>I-A</td>
<td>Class-Level of Evidence</td>
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<tr>
<td>Diagnosis in patients with dyspnea (ambulatory)</td>
<td>I-A</td>
<td>Class-Level of Evidence</td>
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<tr>
<td>Prognosis in patients with known HF (acute)</td>
<td>I-A</td>
<td>Class-Level of Evidence</td>
</tr>
<tr>
<td>Prognosis in patients with known HF (ambulatory)</td>
<td>I-A</td>
<td>Class-Level of Evidence</td>
</tr>
<tr>
<td>Achieving guideline-directed medical therapy (ambulatory)</td>
<td>IIa-B</td>
<td>Class-Level of Evidence</td>
</tr>
<tr>
<td>Natriuretic peptide-guided therapy for chronic HF</td>
<td>IIb-B</td>
<td>Class-Level of Evidence</td>
</tr>
</tbody>
</table>

Class I: conditions for which there is evidence and/or general agreement that a given procedure or treatment is useful and effective; Class IIa: weight of evidence/opinion is in favor of usefulness/efficacy; Class IIb: usefulness/efficacy is less well established by evidence/opinion. Level of Evidence: A: recommendation on the basis of evidence from multiple randomized trials or meta-analyses; Level of Evidence; B: recommendation on the basis of evidence from a single randomized trial or nonrandomized studies.

ACC/AHA = American College of Cardiology/American Heart Association; HF = heart failure.
degree of HF (14), and these findings are not driven by enhanced BNP clearance mediated by natriuretic peptide clearance receptors (15).

**CURRENT USES OF NATRIURETIC PEPTIDES**

A large body of evidence supports the use of the natriuretic peptides (both BNP and NT-proBNP) for the evaluation and management of patients with HF in a variety of clinical settings. Table 1 shows current indications for natriuretic peptide measurements in HF from the American College of Cardiology/American Heart Association (ACC/AHA) guidelines (16). The use of natriuretic peptides in risk stratification has been studied in other clinical settings, such as in acute coronary syndromes, atrial fibrillation, and valvular heart disease. However, there is insufficient evidence to date for a strong recommendation for natriuretic peptide testing in any of these disease states, and none have Level 1 recommendations in the relevant clinical practice guidelines. The evidence supporting specific clinical uses of natriuretic peptides in HF is briefly discussed later.

**DIAGNOSIS IN PATIENTS WITH UNCLEAR SYMPTOMS**

The diagnosis of HF in patients presenting with dyspnea in either urgent care or ambulatory settings is among the strongest indications for natriuretic peptide testing, with a Class I/Level of Evidence: A recommendation in the ACC/AHA guidelines. In the urgent care/emergency setting, the landmark Breathing Not Properly trial evaluated 1,586 patients presenting to the emergency department with unexplained breathlessness, and demonstrated that a BNP threshold of 100 pg/ml could distinguish between a diagnosis of HF and other causes of dyspnea with a high degree of accuracy (area under the receiver-operating characteristic curve: 0.91) (5). In particular, the negative predictive value of BNP <100 pg/ml was high, suggesting that such patients had a very low likelihood of having acute HF as an explanation for their symptoms. Similar data with NT-proBNP from the PRIDE (ProBNP Investigation of Dyspnea) study support the accuracy of NT-proBNP for diagnosis in this same setting (6), although specific age-related thresholds (NT-proBNP >450 pg/ml for patients <50 years of age and NT-proBNP >900 pg/ml for patients 50 years of age or older) have been proposed for NT-proBNP, rather than a single cutpoint. As with all diagnostic tests, interpretation of the results must take into account important sources of false negatives (e.g., obesity), other conditions that may elevate natriuretic peptide levels (e.g., aging and chronic kidney disease), and alternative diagnoses (e.g., acute coronary syndrome or pulmonary embolus). In the setting of “grey zone,” natriuretic peptide values (i.e., intermediate between the optimized threshold to exclude HF and the optimized threshold to definitively diagnose it), other ancillary clinical information and knowledge of prior natriuretic peptide values may be extremely helpful.

Both BNP and NT-proBNP have also been evaluated for use in the outpatient setting in establishing the diagnosis of HF. Importantly, because patients in the ambulatory setting are typically less symptomatic and are less hemodynamically decompensated, diagnostic thresholds are generally lower to avoid false negative results. As with diagnosis of HF in the acute setting, published data suggest a single threshold for BNP, but age adjustment for NT-proBNP. A staged approach to ambulatory patients with suspicion of HF using initial natriuretic peptide screening, followed by echocardiography in patients with elevated values, is supported by the most recent European Society of Cardiology guidelines (17).

**ESTIMATING PROGNOSIS IN PATIENTS WITH KNOWN HF**

The natriuretic peptides are among the most powerful prognostic markers in all forms of clinical HF, including HF with reduced ejection fraction (18), HF with preserved ejection fraction (19), and acute HF (20). In the chronic setting, some of the most compelling data come from the extensive analysis of the Val-HeFT database, where both baseline values and changes over 3 months in BNP and NT-proBNP have been shown to be among the most powerful predictors of future outcomes (21). Similarly, in the acute setting, both elevations of the initial value at the time of presentation (20) and changes with therapy (or value at hospital discharge) are strong predictors of short- (22) and long-term outcomes (23). Although establishing prognosis may be useful for risk stratification, estimating risk alone is of marginal clinical value unless it affects the actual delivery of care in a meaningful way. There has therefore been substantial interest in the concept of titrating or “guiding” clinical care using natriuretic peptide values, so-called biomarker-guided therapy.

**NATRIURETIC PEPTIDE-GUIDED THERAPY**

The concept of guiding chronic care on the basis of levels of a biomarker is common in many chronic diseases, such as diabetes, hypertension, and hepatitis C. The concept of adjusting chronic HF therapy to
A specific natriuretic peptide level has been tested in a variety of small clinical trials, with varying designs and results, and was recently reviewed (24). Although pooled analyses of these studies indicate a 20% to 25% reduction in mortality with biomarker-guided therapy, generalizability has been limited by the small size of the studies, as well as significant heterogeneity in the inclusion criteria, treatment strategies, natriuretic peptide cutpoints, and results (25,26). In light of this uncertainty, current guidelines do not recommend the use of serial natriuretic peptide measurements to guide titration of therapy in chronic HF, and a larger National Institutes of Health trial testing this concept is ongoing (27). It is important to recognize that knowledge of incremental prognostic value may not guarantee the feasibility or ultimate benefit of intensifying drug therapy according to specific biomarker targets.

In addition to guiding therapy of patients with known HF, there is increasing interest in using natriuretic peptide measurements to target at-risk populations without clinical HF for prevention strategies. Two recent randomized clinical trials, PONTIAC (NT-proBNP Guide of Primary Prevention of Cardiovascular Events in Diabetic Patients) (28) and STOP-HF (Screening to Prevent Heart Failure) (29), have demonstrated improvements in clinical outcomes with a strategy of using natriuretic peptide to...
identify high-risk patients for more aggressive HF prevention strategies. Whether and how these promising initial results can be implemented on a broader population level is an area of active ongoing research.

REFINEMENTS OF CLINICAL NATRIURETIC PEPTIDE TESTING

When natriuretic peptide testing was first introduced for clinical use more than 15 years ago, it was reassuring that levels were invariably elevated in most settings of HF (especially during exacerbations) and often tracked with clinical stability. However, clinicians also realized that levels may vary widely among patients with the same degree of symptoms or echocardiographic parameters, which requires occasional adjustments to individualize clinical interpretation. For example, almost a quarter of patients with chronic symptomatic HF may have natriuretic peptide levels in the lower ranges (30), and their levels should be explained by other contributing factors (e.g., obesity, ischemia) when clinical presentation is out of proportion to their degree of disease severity by other measures. In contrast, euvoletic patients (particularly older women, likely with lower lean mass) may have higher ranges (13). Meanwhile, natriuretic peptide levels are consistently higher in patients with underlying chronic kidney disease. As patients reach end-stage renal disease, natriuretic peptide levels can be 10-fold higher than the operational range common to non-end-stage renal disease patients (31). Yet, in those undergoing hemodialysis, natriuretic peptides may not track tightly with volume excess or insufficient clearance, but an increase over time still portends poor prognosis (32). Taken together, tracking individual patients’ own natriuretic peptide trajectories seems to be the prevailing clinical strategy, integrating the results by accounting for different factors (33). It should be emphasized that patients with an elevated natriuretic peptide level, even in the absence of ongoing acute symptoms, have a poor prognosis.

Analytical variability has long been an issue with point-of-care BNP testing, and may differ from lot to lot. Laboratory-based testing has improved this variability, but different antibodies used by different assays from different vendors may affect diagnostic ranges and are often difficult to harmonize. Furthermore, the intrinsic biological variability appeared significant for serial measurements of both BNP and NT-proBNP—up to 130% reported in one study (34). This clearly can have an impact on the overall accuracy of the results.

“WET” AND “DRY” NATRIURETIC PEPTIDE LEVELS

The rise in natriuretic peptide levels was originally explained by an acute increase in myocardial stretch, leading to transcriptomic expression and rapid induction of the counter-regulatory machinery to maintain homeostasis. However, over time, more constitutively produced (“dry”) natriuretic peptides appear, and congestion-driven (“wet”) natriuretic peptide levels often comprise a portion of the total quantity of detectable natriuretic peptide levels. This may explain why, in the advanced HF setting, changes in natriuretic peptides may not necessarily track with blood volume or invasive hemodynamic measurements, both cross-sectionally and over time (35). Therefore, any rise in natriuretic peptide levels requires careful interpretation in clinical context, as intensifying medical therapy cannot relieve all factors affecting natriuretic peptide levels. Unexplained and persistent rises in natriuretic peptide levels may warrant thorough investigations (e.g., occult arrhythmias, dietary indiscretion, systemic diseases [e.g., thyroid dysfunction or cancer]).

This intricate balance between wet and dry natriuretic peptides is further illustrated by changes in natriuretic peptide levels following initiation of drug therapies. For example, early initiation of antiadrenergic blockers may paradoxically raise natriuretic peptide levels (36), whereas long-term benefits of antiadrenergic therapy may track with overall reduction in natriuretic peptide levels over time (37). More recently, in the PARADIGM-HF trial, administration of sacubitril/valsartan has been noted to block neprilysin’s ability to degrade endogenous BNP, thus leading to an overall increase in BNP, yet its beneficial effects resulted in a reduction in NT-proBNP levels (38) (Central Illustration). There has been a recent suggestion that angiotensin receptor-neprilysin inhibitor may alter glycosylation and deglycosylation of NT-proBNP, but the precise role that this plays in the interpretation of natriuretic peptide levels for patients taking sacubitril/valsartan is speculative (39). These observations highlight the importance of taking the biological underpinnings of the disease and its treatment effects into account when interpreting natriuretic peptide levels.

NATRIURETIC PEPTIDE TESTING IN ACUTE HF

Beyond its diagnostic role, the usefulness of natriuretic peptide-guided therapy for acutely decompenated HF is less well established (16). In fact,
NT-proBNP seems to be more affected by HF than BNP, in part due to the relatively longer half-life of NT-proBNP, which may not equilibrate to steady-state levels until up to 1 week after clinical stabilization. Transient fluctuations in renal functions, as a result of decongestive and vasoactive therapies during decompensated states, may also influence renal clearance of natriuretic peptides. This common “rebound” scenario, observed in some cases after the initial aggressive diuresis phase, results in fluctuating renal volume status in the transition to clinical stability (40). Hence, the lack of immediate reductions in natriuretic peptides following decongestion should not necessarily concern clinicians and their patients. Yet, because lower levels often track with better outcomes, further investigations are warranted if levels become persistently elevated.

On the contrary, clinicians often ignore the critical period during HF hospitalizations where risk stratification can be crucial in planning for post-discharge care. Although the >30% reduction in natriuretic peptide levels has been considered an unofficial therapeutic goal, it appears that the overall ranges of natriuretic peptide elevation (especially in the chronic setting [i.e., “dry” natriuretic peptide]) may provide more prognostic value than acute changes. In other words, the persistence of high (>5,000 pg/ml) NT-proBNP levels long after decongestion portends poor outcomes, regardless of relative changes. This point is often overlooked, but is clearly illustrated in the post-hoc analysis of the PROTECT (ProBNP Outpatient Tailored Chronic Heart Failure Trial) trial (41). It points to the important role of natriuretic peptide testing for risk stratification, not only in terms of predicting rehospitalization or mortality, but in the management of patient expectations, including advanced therapies or end-of-life care considerations. It is reasonable to measure natriuretic peptide levels at the time a patient is admitted to the hospital for HF and during an early outpatient follow-up visit.

**CONCLUSIONS**

There is little debate that natriuretic peptide testing has revolutionized clinical care for patients with HF over the past decade. The measurement of natriuretic peptides can help us to gain insights into the underlying biology of the HF syndrome. These biomarkers now have the highest level of recommendation in the most recent ACC/AHA guidelines for both the diagnosis of patients with uncertain symptoms, and for establishing prognosis in patients with established HF. However, clinicians need to appreciate the complex dynamics of natriuretic peptide levels, taking patient characteristics, comorbidities, and other therapies into account as they use and interpret these biomarkers. Ongoing studies on the use of natriuretic peptide measurements may help us to individualize our treatment plans above and beyond guideline recommendations.

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41. Gaggin HK, Truong QA, Rehman SU, et al. Characterization and prediction of natriuretic peptide “nonresponse” during heart failure management: results from the ProBNP Outpatient Tailored Chronic Heart Failure (PROTECT) and the NT-proBNP-Assisted Treatment to Lessen Serial Cardiac Readmissions and Death (BATTLESCARRED) study. Congest Heart Fail 2013;19:135–42.

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