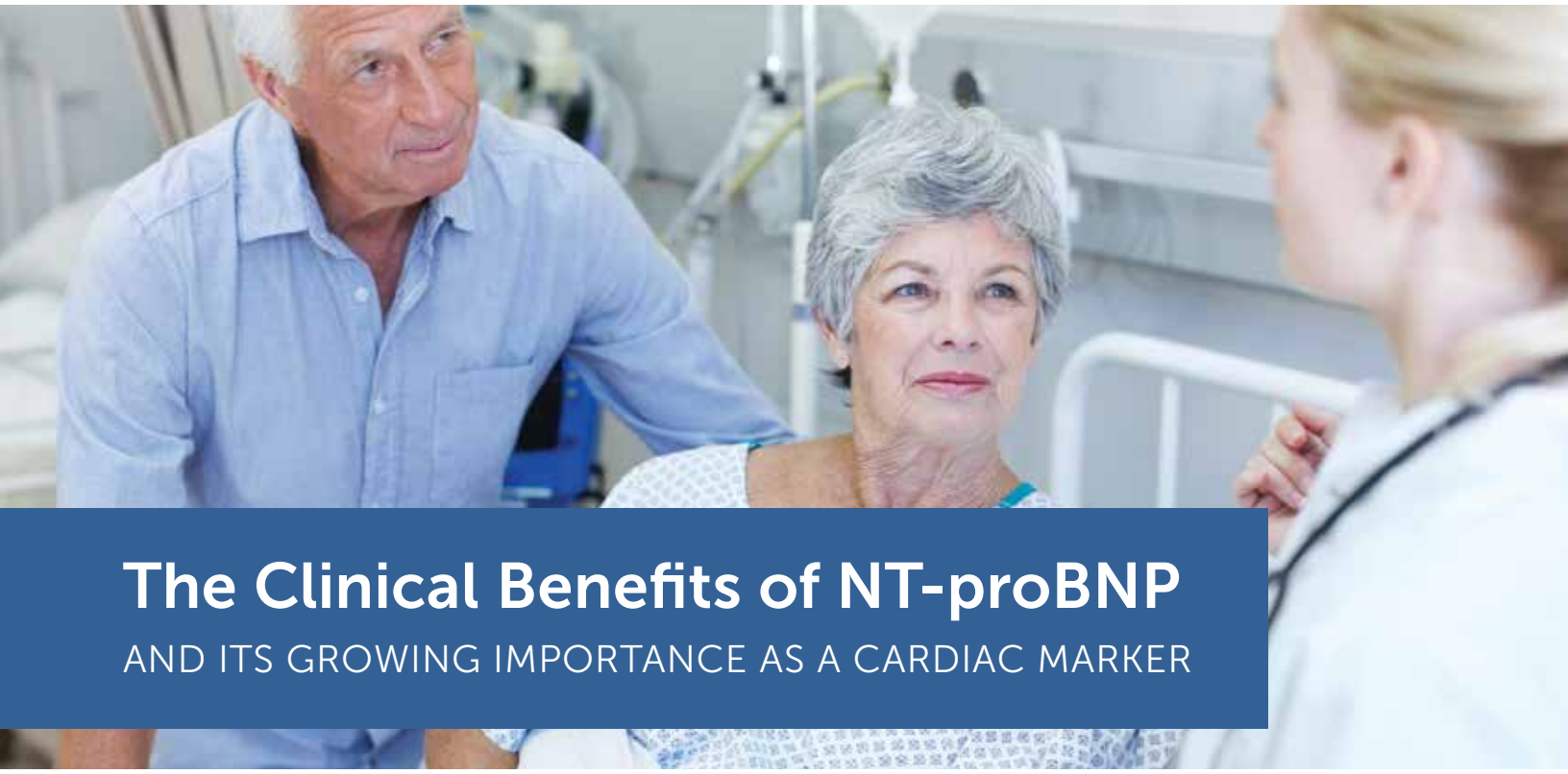


The Clinical Benefits of NT-proBNP

AND ITS GROWING IMPORTANCE AS A CARDIAC MARKER

Heart Failure and the Role of Natriuretic Peptides

Heart Failure (HF) is a complex health challenge of rising global concern affecting 5.1 million Americans and 23 million individuals worldwide.[1,2] This progressive condition is characterized by weakening of the heart muscle and inefficient blood circulation to the body. HF is currently the only major cardiovascular disease that is still increasing in incidence and prevalence, continuing to escalate due to the aging population and improved survival rates after heart attack.

HF carries a substantial financial burden as the mortality rate of this condition is staggering, with roughly 50% of people diagnosed with HF dying within 5 years.[3] Consequently, HF is a leading cause of hospitalizations and readmissions, with 1 million discharges annually in the US and approximately 1 in 4 HF patients being readmitted within 30 days of discharge.[1,4] With this resource utilization comes an overall annual spend of $32 billion dollars towards HF in the US and 1-2% of all health care costs in developed nations.[1,5]

As healthcare policy evolves, more focus is placed on improving patient outcomes and resource utilization for chronic conditions like HF. Timely diagnosis and effective treatment is vital for improvement. **NT-proBNP and BNP** have emerged as promising markers to aid in the diagnosis of HF, assess patient prognosis, and aid in guiding treatment.

Benefits of NT-proBNP

Implementation of NT-proBNP testing for patients with Acute Decompensated Heart Failure has been shown to decrease hospital length of stay, rehospitalizations, and improve rates of morbidity and mortality, presumably due to earlier diagnosis and treatment.[6]

NT-proBNP and BNP as HF Biomarkers

The natriuretic peptides NT-proBNP and BNP result from cleavage of proBNP. ProBNP is secreted by heart muscle cells in response to increased myocardial wall stretch that occurs with increased circulating blood volume (as in the case of HF). BNP the biologically active peptide causes blood vessel expansion (vasodilation), increased urine production (diuresis), and excretion of sodium in the urine (natriuresis), to decrease blood volume and increase cardiac output.

International guidelines highlight the clinical utility of these biomarkers

Diagnosis or Exclusion of HF

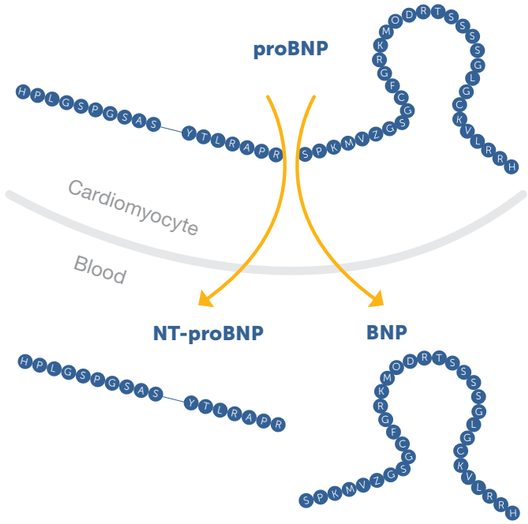
- To support clinical decision making for the diagnosis of HF in ambulatory patients with dyspnea (shortness of breath), especially in the setting of clinical uncertainty.[9-12]
- To support clinical judgment for the diagnosis of acutely decompensated HF among patients presenting to the ED with acute dyspnea.[9-11]

Prognosis of HF

- For establishing prognosis or disease severity in patients with chronic HF or acutely decompensated HF.[9-12]

Achieve GDMT
(guideline-directed medical therapy)

- To achieve optimal dosing of GDMT in select ambulatory patients followed in a well-structured HF disease management program.[9]



Distinctive Features of NT-proBNP and BNP[7]

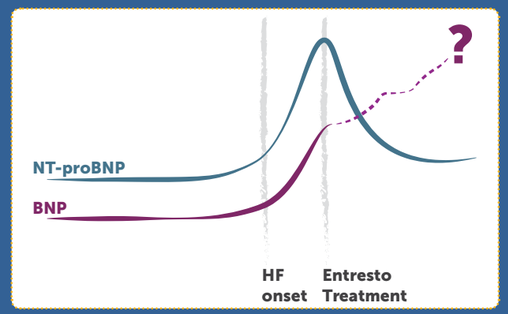
NT-proBNP	BNP
Biologically inactive	Biologically active
76 amino acids	**32** amino acids
Half-Life: **~120 min**	Half-Life: **~22 min**
Plasma Stability*: **up to 3 days**	Plasma Stability*: **~4 hrs**

*EDTA Plasma Stability at Room Temperature

Entresto™ and BNPs

Entresto, the newly FDA approved drug for HF[13], disrupts BNP levels however **NT-proBNP** "remains a useful cardiac biomarker to assess therapeutic effect and prognosis in patients treated with [the drug]."[14]

Entresto is a trademark of Novartis AG.



Why choose NT-proBNP?

As the use of natriuretic peptide testing has increased for HF, more research has been conducted focusing on the effectiveness of utilizing NT-proBNP and BNP. Studies have shown that NT-proBNP has several clear advantages with respect to its stability, harmonization with other methods, and clinical performance.

Biochemical Stability

- The longer half-life of NT-proBNP (approx. 120 minutes), in contrast to BNP (approx. 22 minutes) allows for measurement of steady-state ventricular function that is less subject to short-term physiologic variations.[7]
- NT-proBNP is less prone to degradation *in vivo* and *in vitro*, allowing for more stable blood specimens for testing.[8] In contrast, BNP has demonstrated poor *in vitro* stability in several studies, a definite concern if testing is not performed shortly after specimen collection.[15,16]

Assay Standardization

- All licensed NT-proBNP tests provide comparable results as they are calibrated to the Roche method, resulting in harmonization across different laboratories and testing platforms.[7]
- In contrast, BNP tests are not harmonized to a single method and multiple antibodies and targets are utilized. As a result, different BNP testing platforms may yield different results.[7]

Proven Clinical Performance

- NT-proBNP has been proven to be superior to BNP for predicting mortality, morbidity and hospitalization for HF.[17]
- NT-proBNP has been shown to have better accuracy than BNP for identifying mild HF, as shown in a study examining the diagnostic accuracy of BNP and NT-proBNP for a wide severity range of HF patients, including a healthy control group.[18]
- The use of NT-proBNP to guide pharmacological therapy in patients with chronic HF is associated with reduced all-cause mortality and HF-related hospitalization, while the use of BNP-guided therapy is not significantly associated with reduced mortality and morbidity.[19]
- A single BNP cutpoint of 100 pg/mL for ruling out HF in the acute setting has been shown to provide a poor Negative Predictive Value (NPV = 89%).[20] In a study of outpatients with chronic, stable systolic HF, 1 in 5 symptomatic patients with HF tested within the normal range for BNP (<100 pg/mL), raising doubt for the utilization of a single cutpoint in the acute and outpatient setting.[21]

NT-proBNP Utilization and Interpretation

Heart failure is a complex clinical syndrome and patients can often present with non-specific symptoms often shared by other disease states, making diagnosis a challenge. NT-proBNP has emerged as a useful tool to aid physicians in this diagnosis. Studies have provided optimal cutpoints for NT-proBNP to help physicians confidently utilize this assay for the exclusion and diagnosis of HF in the acute and outpatient setting.

Each laboratory should investigate the transferability of the expected values to its own patient population and, if necessary, determine its own reference ranges.

Exclusion of Heart Failure

The greatest value of natriuretic peptide testing is its ability to confidently rule out HF. Optimal cutpoints would produce a Negative Predictive Value of 100%, ensuring the risk for overlooking a patient with HF is minimized.

ACUTE SETTING

Patient Population	Optimal Cutpoint	Sensitivity	Negative Predictive Value
All ages	300 ng/L	99%	98-99%

A single cutpoint can be utilized for dyspnoeic patients presenting in the acute setting to rule out acute HF, providing optimal sensitivity and Negative Predictive Values.[22, 23]

OUTPATIENT SETTING

Patient Population	Optimal Cutpoint	Negative Predictive Value
< 75 years	125 ng/L	100%
≥ 75 years	450 ng/L	100%

In the ambulatory setting, age stratified cutpoints can be utilized to effectively rule out chronic HF.[24]

Can NT-proBNP be used in other settings?

1

ASSESSMENT IN PATIENTS WITH KIDNEY FAILURE

The presence of kidney failure can result in higher NT-proBNP and BNP results due to the reduction in renal clearance of both peptides. Studies show that:

- NT-proBNP is a superior predictor of heart failure and mortality in patients with kidney disease.[25, 26]
- In hemodialysis patients, changes in NT-proBNP values are significant risk predictors.[27]

2

CARDIOVASCULAR RISK ASSESSMENT AND PROGNOSIS

- NT-proBNP levels independently predict HF and cardiovascular death in older adults. NT-proBNP levels frequently change over time and these fluctuations reflect dynamic changes in cardiovascular risk.[28]
- In hospitalized patients with HF, measurement of NT-proBNP levels at discharge may provide useful prognostic information, as a failure of NT-proBNP levels to fall is a poor prognostic sign, suggesting more intensive monitoring or treatment may be necessary.[7, 29]

3

SYMPTOM SEVERITY ASSESSMENT

- There is a significant relationship between New York Heart Association (NYHA) HF symptom severity and NT-proBNP levels.
- As symptom severity rises so too do the median NT-proBNP levels.[22]

Aid in Diagnosis of HF

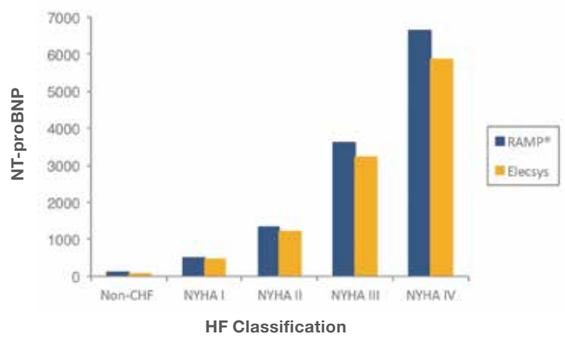
The PRIDE and ICON contemporary NT-proBNP studies have demonstrated optimal age stratified cutpoints for the diagnosis of acute heart failure among dyspnoeic patients. Three age stratified cutpoints provide optimal accuracy, while maintaining ease of use (450/900/1800 ng/L for ages <50/50-75/>75 years).[22,23]

NT-proBNP at the Point of Care

- In the Emergency Department setting, it may be beneficial to have NT-proBNP turn-around times less than 60 minutes to optimally aid physicians in the diagnosis or exclusion of HF in acutely dyspnoeic patients.
- The use of a whole blood, rapid Point of Care device can improve the utility of NT-proBNP in the Emergency Department setting.

Point of Care

Median NT-ProBNP According to Severity of Heart Failure[30]



Making the switch to NT-proBNP

Interpreting NT-proBNP is as simple as interpreting BNP results

With education and training, incorporating NT-proBNP into your facility's protocols can provide optimal benefits for patients and improve utility for physicians.

KEY FEATURES TO NOTE

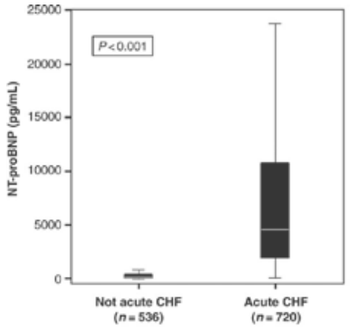
- NT-proBNP levels are typically 2-10 times higher than BNP levels in patients with HF.[31]
- NT-proBNP results provide optimal differentiation between patient populations with acute HF and those without, allowing for accurate result interpretation as shown in the ICON study.

Non-Heart Failure Related Changes in Natriuretic Peptides

Caution should be utilized when interpreting **both BNP and NT-proBNP** results as there are a number of non-HF conditions that can cause increases and decreases in these biomarkers.

- Both BNP and NT-proBNP have high serum levels in the presence of advanced age, female gender, anemia, and renal dysfunction.[7, 32]
- High levels of serum natriuretic peptides can have causes other than heart failure including: left ventricular hypertrophy, ischemia, tachycardia, right ventricular overload, hypoxemia (including pulmonary embolism), sepsis, chronic obstructive pulmonary disease (COPD), diabetes, and cirrhosis of the liver.[12]
- Obesity or treatment with diuretics, angiotensin-converting enzyme (ACE) inhibitors, beta-blockers, angiotensin II receptor antagonists (ARBs) and aldosterone antagonists can reduce levels of serum natriuretic peptides.[12]

NT-proBNP Values Between Diagnostic Groups In the ICON Study[22]



Diagnostic category	Median NT-proBNP	IQR
Not acute CHF	108 pg/mL	37–381 pg/mL
Acute CHF	4639 pg/mL	1882–10818 pg/mL

INCREASES

- Advanced Age
- Female Gender
- Anemia
- Renal Dysfunction
- Left Ventricular Hypertrophy
- Ischemia
- Tachycardia
- Right Ventricular Overload
- Hypoxemia (including pulmonary embolism)
- Sepsis
- Chronic Obstructive Pulmonary Disease (COPD)
- Diabetes
- Cirrhosis of the Liver

DECREASES

- Obesity
- Treatment with Diuretics
- Angiotensin-converting enzyme (ACE) inhibitors
- Beta-blockers
- Angiotensin II receptor antagonists (ARBs)
- Aldosterone antagonists

RAMP® NT-proBNP

Healthcare professionals require a diagnostic testing solution with built-in process controls to provide optimal result quality and improve the speed of diagnosis.

Speed

- Testing performed on EDTA whole blood
- Results available in approx. 15 minutes
- Optimized patient management with testing available in a variety of settings
- No user calibration or maintenance required

Precision

- Central laboratory quality results comparable to the Roche proBNP assay in terms of imprecision, linearity, lower detection limit, and functional sensitivity

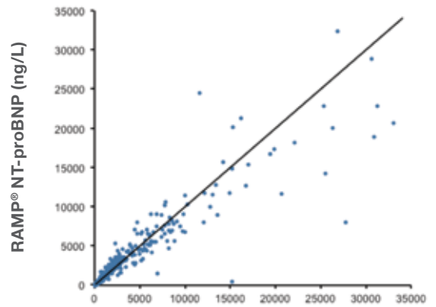
Accuracy

- Excellent correlation to the Roche proBNP test
- RAMP® tests utilize the **proprietary RAMP® Ratio** that corrects for potential sources of variability:
  - Operator technique
  - Sample variability
  - Environmental conditions

Test Summary	
Time to Result	Approx. 15 minutes
Sample Type	EDTA Whole Blood
Sample Volume	75 µL
Sample Stability	2 hrs Room Temperature 48 hrs Refrigerated (2-8°C)
Reportable Range	27 - 22,000 ng/L (USA) 5 - 35,000 ng/L (outside USA)
Calibration Frequency	No user calibration

Laboratory Quality Performance

RAMP® = 1.01Elecsys + 14.6 **R = 0.98**



RAMP® 200

- A high throughput, multi-port laboratory reader
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References

1. Go AS, Mozaffarian D, Roger VL, et al. Heart disease and stroke statistics--2013 update: a report from the American Heart Association. Circulation. 2013;127(1):e6–e245. doi:10.1161/CIR.0b013e31828124ad.
2. McMurray JJ, Petrie MC, Murdoch DR, Davie AP. Clinical epidemiology of heart failure: public and private health burden. Eur Heart J. 1998;19 Suppl P:P9–P16.
3. Roger VL, Weston SA, Redfield MM, et al. Trends in heart failure incidence and survival in a community-based population. JAMA. 2004;292:344–350. doi:10.1016/j.accreview.2004.11.055.
4. Krumholz HM, Merrill AR, Schone EM, et al. Patterns of hospital performance in acute myocardial infarction and heart failure 30-day mortality and readmission. Circ Cardiovasc Qual Outcomes. 2009;2:407–413. doi:10.1161/CIRCOUTCOMES.109.883256.
5. Liao L, Allen LA, Whellan DJ. Economic burden of heart failure in the elderly. Pharmacoeconomics. 2008;26:447–462.
6. Green SM, Redmond P, Januzzi JL, et al. The impact of amino-terminal pro-brain natriuretic peptide testing on hospital length of stay and morbidity in patients with acute decompensated heart failure. Arch Pathol Lab Med. 2007;131:473–476.
7. Melanson SEF, Lewandrowski EL. Laboratory testing for B-type natriuretic peptides (BNP and NT-proBNP): clinical usefulness, utilization, and impact on hospital operations. Am J Clin Pathol. 2005;124 Suppl:S122–S128.
8. Clerico A, Emdin M. Diagnostic accuracy and prognostic relevance of the measurement of cardiac natriuretic peptides: a review. Clin Chem. 2004;50:33–50. doi:10.1373/clinchem.2003.024760.
9. Yancy CW, Jessup M, Bozkurt B, Butler J, Casey D E J. 2013 ACCF/AHA Guideline for the Management of Heart Failure: A Report of the American College of Cardiology Foundation/American Heart Association Task Force …. Circulation. 2013. Available at: http://www.ncbi.nlm.nih.gov/pubmed/23741058.
10. McMurray JJ V, Adamopoulos S, Anker SD, et al. ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure 2012: The Task Force for the Diagnosis and Treatment of Acute and Chronic Heart Failure 2012 of the European Society of Cardiology. Developed in collaboration with the Heart. Eur Heart J. 2012;33(14):1787–847. doi:10.1093/eurheartj/ehs104.
11. Christenson RH, Apple FS, Cannon CP, College M. The National Academy of Clinical Biochemistry GUIDELINES BIOMARKERS OF ACUTE CORONARY SYNDROMES AND. 2007.
12. National Collaborating Centre for Acute and Chronic Conditions. Chronic heart failure. Management of chronic heart failure in adults in primary and secondary care. London (UK):National Institute for Health and Clinical Excellence (NICE); 2010 Aug. 45 p. (Clinical guideline; no. 108).
13. Novartis' new heart failure medicine LCZ696, now called Entresto(TM), approved by FDA to reduce risk of cardiovascular death and heart failure hospitalization. Novartis Press Release. 07 July 2015.
14. Langenickel TH, Dole WP. Angiotensin receptor-neprilysin inhibition with LCZ696: a novel approach for the treatment of heart failure. Drug Discovery Today: Therapeutic Strategies. Vol. 9, No. 4, 2012; e131-e139.
15. Shimizu H, Aono K, Masuta K, Asada H, Misaki A, Teraoka H. Stability of brain natriuretic peptide (BNP) in human blood samples. Clin Chim Acta. 1999;285:169–172. doi:10.1016/S0009-8981(99)00112-6.
16. Shimizu H, Aono K, Masuta K, Asada H, Misaki A, Teraoka H. Degradation of human brain natriuretic peptide (BNP) by contact activation of blood coagulation system. Clin Chim Acta. 2001;305:181–186.
17. Masson S, Latini R, Anand IS, et al. Direct comparison of B-type natriuretic peptide (BNP) and amino-terminal proBNP in a large population of patients with chronic and symptomatic heart failure: the Valsartan Heart Failure (Val-HeFT) data. Clin Chem. 2006;52:1528–1538. doi:10.1373/clinchem.2006.069575.
18. Emdin M, Passino C, Prontera C, et al. Comparison of brain natriuretic peptide (BNP) and amino-terminal ProBNP for early diagnosis of heart failure. Clin Chem. 2007;53:1289–1297. doi:10.1373/clinchem.2006.080234.
19. Savarese G, Trimarco B, Dellegrottaglie S, et al. Natriuretic peptide-guided therapy in chronic heart failure: a meta-analysis of 2,686 patients in 12 randomized trials. PLoS One. 2013;8:e58287. doi:10.1371/journal.pone.0058287.
20. Maisel AS, Krishnaswamy P, Nowak RM, et al. Rapid measurement of B-type natriuretic peptide in the emergency diagnosis of heart failure. N Engl J Med. 2002;347:161–167. doi:10.1056/NEJMoa020233.
21. Tang WHW, Girod JP, Lee MJ, et al. Plasma B-type natriuretic peptide levels in ambulatory patients with established chronic symptomatic systolic heart failure. Circulation. 2003;108:2964–2966. doi:10.1161/01.CIR.0000106903.98196.B6.
22. Januzzi JL, van Kimmenade R, Lainchbury J, et al. NT-proBNP testing for diagnosis and short-term prognosis in acute destabilized heart failure: an international pooled analysis of 1256 patients: the International Collaborative of NT-proBNP Study. Eur Heart J. 2006;27:330–337. doi:10.1093/eurheartj/ehi631.
23. Januzzi JL, Camargo CA, Anwaruddin S, et al. The N-terminal Pro-BNP investigation of dyspnea in the emergency department (PRIDE) study. Am J Cardiol. 2005;95:948–954. doi:10.1016/j.amjcard.2004.12.032.
24. Gustafsson F, Steensgaard-Hansen F, Badskjær J, Poulsen AH, Corell P, Hildebrandt P. Diagnostic and Prognostic Performance of N-Terminal ProBNP in Primary Care Patients With Suspected Heart Failure. J Card Fail. 2005;11(5):S15–S20. doi:10.1016/j.cardfail.2005.04.022.
25. Jafri L, Kashif W, Tai J, et al. B-type natriuretic peptide versus amino terminal pro-B type natriuretic peptide: selecting the optimal heart failure marker in patients with impaired kidney function. BMC Nephrol. 2013;14(November 2012):117. doi:10.1186/1471-2369-14-117.
26. deFilippi CR, Seliger SL, Maynard S, Christenson RH. Impact of renal disease on natriuretic peptide testing for diagnosing decompensated heart failure and predicting mortality. Clin Chem. 2007;53:1511–1519. doi:10.1373/clinchem.2006.084533.
27. Pastural-Thaunat M, Ecochard R, Boumendjel N, et al. Relative Change in NT-proBNP Level: An Important Risk Predictor of Cardiovascular Congestion in Haemodialysis Patients. Nephron Extra. 2012;2(1):311–8. doi:10.1159/000343897.
28. deFilippi CR, Christenson RH, Gottdiener JS, Kop WJ, Seliger SL. Dynamic Cardiovascular Risk Assessment in Elderly People. The Role of Repeated N-Terminal Pro-B-Type Natriuretic Peptide Testing. J Am Coll Cardiol. 2010;55:441–450. doi:10.1016/j.jacc.2009.07.069.
29. O'Brien RJ, Squire IB, Demme B, Davies JE, Ng LL. Pre-discharge, but not admission, levels of NT-proBNP predict adverse prognosis following acute LVF. Eur J Heart Fail. 2003;5:499–506. doi:10.1016/S1388-9842(03)00098-9.
30. Lee-Lewandrowski E, Januzzi JL, Green SM, et al. Multi-center validation of the Response Biomedical Corporation RAMP® NT-proBNP assay with comparison to the Roche Diagnostics GmbH Elecsys® proBNP assay. Clin Chim Acta. 2007;386:20–24. doi:10.1016/j.cca.2007.07.015.
31. Hall C. Essential biochemistry and physiology of (NT-pro)BNP. Eur J Heart Fail. 2004;6:257–260. doi:10.1016/j.ejheart.2003.12.015.
32. Novaes M, Curiati C, Silvestre OM, et al. Agreement of BNP and NT-proBNP and the influence of clinical and laboratory variables. Einstein. 2013;11:273–277.

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