Implications of serial measurements of natriuretic peptides in heart failure

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Implications of serial measurements of natriuretic peptides in heart failure: insights from BIOSTAT-CHF

Natriuretic peptides [NP, including B-type natriuretic peptide (BNP) and amino-terminal prohormone of BNP (NT-proBNP)] are the gold-standard biomarkers in heart failure (HF) management,[1] with NP levels at presentation/admission routinely used for diagnostic and prognostic purposes.[2] NP levels at discharge/follow-up also show association with outcomes,[3] and NP levels following HF treatment add further value to tailoring risk.[4] However, the usefulness of NP serial measurements beyond conventional HF treatment in clinical practice still remains a matter of controversy.[3,5] A cohort with current HF guideline-based treatment would provide an ideal setting to revisit usefulness of NP serial measurements in risk stratification of HF patients, including the role of recently identified BNP molecular forms.[6] The European multi-national BIOlogy Study to TAilored Treatment in Chronic Heart Failure (BIOSTAT-CHF) provides an opportunity for the aforementioned analysis, being a European cohort in which serial sampling of NPs was done before and after titration of HF medications according to current European guidelines in a multi-centre, observational, real-world setting.[7]

The aims of the present study were to investigate the association with HF outcomes, effects of HF guideline treatment, and the implications of NP serial measurement in the BIOSTAT-CHF cohort.[7]

From the total cohort, 757 patients with available plasma samples at baseline (V1) and at follow-up (V2, approximately 9 months apart) were measured for BNP and BNP molecular form, BNP 5–32[6] (see *Table 1* for methods on measurements). NT-proBNP measurement was only available at baseline (V1); therefore, analyses related to this peptide were limited to baseline (V1). The primary endpoints were all-cause mortality and a composite of mortality with HF rehospitalisation (mortality/HF) at 3 years, and overall from baseline (V1). Changes in dosage titrations and response of peptide levels were investigated by splitting the population into two groups based on treatment up-titration, as previously reported (see online supplementary material).[7]

Demographics and clinical measurements are described in *Table 1*. At baseline (V1), NP levels (BNP, NT-proBNP, and BNP 5–32) were strongly correlated with each other ($r_s = 0.635–0.904$, $P < 0.001$). Cox regression modelling showed baseline BNP levels to be associated with mortality [hazard ratio (HR) 1.99, 95% confidence interval (CI) 1.23–3.23; $P = 0.005$] and mortality/HF (HR 1.72, 95% CI 1.25–2.37; $P = 0.001$). NT-proBNP and detection of BNP 5–32 were similarly associated with mortality (HR $\geq$1.85, 95% CI 1.15–3.20; $P \leq 0.012$) and mortality/HF [HR $\geq$1.54, 95% CI 1.14–3.22; $P \leq 0.015$) after adjustment for the BIOSTAT-CHF compact model[7] (*Table 2*). All three NPs retained their associations with outcomes after further adjustment with additional NP confounders (online supplementary *Table S1A*). With regard to the effect of HF treatment, significantly reduced BNP levels were observed only when at least one medication was up-titrated, whereas BNP 5–32 was reduced regardless of drug up-titration (*Table 3*). A general linear model analysis for repeated measures confirmed these findings (online supplementary *Table S1B*). For serial measurements (*Table 4*), when BNP baseline (V1) and follow-up (V2) levels were compared, follow-up (V2) measurements were more strongly associated with all cause-mortality than baseline (V1) (chi-square: 67.1 vs. 16.0). However, even if the combination of baseline (V1) and follow-up (V2) measurements were significant (chi-square: 66.7, $P < 0.001$), there was no added value to the follow-up (V2) measurement alone, as the role of the baseline (V1) measurement was not preponderant ($P = 0.878$). Similarly, follow-up BNP 5–32 measurement showed a stronger association with all-cause mortality than the baseline value (chi-square: 64.3 vs. 18.8); however, the combination of baseline (V1) and follow-up (V2) measurements was significantly better (chi-square: 69.8), with the baseline (V1) level providing additional value (chi-square: 5.5, $P = 0.017$) to follow-up (V2) measurement alone. Furthermore, in patients that did not achieve $\geq$50% dose treatment but still showed BNP 5–32 to decrease from detectable to undetectable levels (or high-low for BNP) exhibited better outcomes than those who displayed increased levels at follow-up (online supplementary *Figures S1* and *S2*).

There are three main findings of the present investigation. Firstly, baseline NP levels were independently associated with adverse outcomes, with comparable results for BNP, NT-proBNP, and BNP 5–32. Secondly, response to HF guideline treatment up-titration was associated with a decrease in both BNP and BNP 5–32 levels. Finally, even if both BNP and BNP 5–32 showed stronger association with all-cause mortality at follow-up measurement compared to baseline, combination of baseline and follow-up measurements did not add value for BNP beyond follow-up alone, whereas BNP 5–32 did.

The recent North American GUIDE-IT trial[8] showed guideline-directed medical therapy (GDMT) guided by NT-proBNP levels was not superior to GDMT alone and that GDMT intensity was associated with lower NT-proBNP levels and further that low NP levels at follow-up (NT-proBNP levels $\leq$1000 pg/mL during GDMT) were associated with better outcomes.[9] Consistent with this, the present study based on a European real-world cohort showed that follow-up values after guideline-based treatment were more associated with outcomes for both BNP and BNP 5–32 (online supplementary *Figures S3* and *S4*). In this context, analysis of the NP response in the BIOSTAT-CHF cohort, with medications optimised according to HF guidelines, confirmed the association of baseline NP levels (BNP, NT-proBNP and BNP 5–32) with adverse outcomes, and follow-up levels after treatment to show better association with adverse outcomes when compared to baseline levels, consistent with previous reports.[1,2,4,10,11] This is in line with a previous finding in another real-world cohort conducted in the UK in which the measurement of follow-up NT-proBNP, after optimisation of pharmacotherapy, although preceding current guidelines, provided more value than baseline measurements alone.[10] The difference in added value of combined

Table 1 Patient characteristics

	Patients with follow-up visit (n = 757)		
	Visit 1	**Visit 2**	***P*-value**
Age, years	69 (60–77)		
Male sex	76%		
Current smoker	14%		
Ischaemic aetiology	54%		
Diabetes mellitus	31%		
COPD	18%		
Previous HF hospitalisation	29%		
NYHA class			<0.001*
I	3%	16%	
II	42%	59%	
III	47%	24%	
IV	8%	1%	
LV ejection fraction (%)	30 (25–36)	35 (28–43)	<0.001*
Pulmonary congestion	49%	11%	<0.001*
Peripheral oedema	49%	24%	<0.001*
Systolic blood pressure (mmHg)	122 (110–140)	123 (110–140)	0.654
Diastolic blood pressure (mmHg)	75 (68–85)	75 (66–80)	0.011*
Heart rate (bpm)	75 (65–88)	70 (61–80)	<0.001*
Beta-blocker	85%	93%	<0.001*
ACEi or ARB	74%	89%	<0.001*
Haemoglobin (g/dL)	13.4 (12.1–14.5)	13.3 (12.1–14.3)	0.030*
Urea (mmol/L)	9.4 (6.8–14.3)	10.3 (7.1–15.7)	<0.001*
eGFR[a] (mL/min/1.73 m^2)	66 (49–82)	61 (46–79)	<0.001*
Sodium (mmol/L)	140 (137–142)	139 (137–142)	0.209
BNP (pg/mL)	202 (85–406)	134 (49–349)	0.001*
NT-proBNP (ng/L)	2236 (971–4654)	–	–
BNP 5–32[a]	50% [0.2 (0–0.5)]	25% [0 (0–0)]	<0.001*
Endpoints			
2 years			
Death	83		
Death/HF	219		
3 years			
Death	97		
Death/HF	230		

ACEi, angiotensin-converting enzyme inhibitor; ARB, angiotensin receptor blocker; BNP, B-type natriuretic peptide; BP, blood pressure; COPD, chronic obstructive pulmonary disease; eGFR, estimated glomerular filtration rate (Chronic Kidney Disease Epidemiology Collaboration formula); HF, heart failure; LV, left ventricular; NT-proBNP, N-terminal pro B-type natriuretic peptide; NYHA, New York Heart Association.

Combined data are shown as median (interquartile range) for continuous variables and as a % for categorical variables. *P*-values for visit 1 vs. visit 2 are quoted for Wilcoxon matched-pair signed-rank tests for continuous variables and McNemar test for categorical variables. BNP 5–32 is reported as a ratio of molecular form signal intensity against an internal reference standard.

BNP was measured using Luminex multiplexed bead-based immunoassays (Alere, San Diego, CA, USA) and validated in a small subset using a commercial assay [RapidPIA®, Sekisui Medical Co.; r^2 = 0.825)]. NT-proBNP measured using the Roche NT-proBNP assay (Roche Diagnostics, Risch-Rotkreuz, Switzerland). BNP 5–32 was measured using matrix-assisted laser desorption ionisation-time of flight-mass spectrometry (MALDI-ToF-MS).[6] BNP 4–32 and BNP 3–32 were also detected in the same assay as BNP 5–32 but were not as sensitive and not comparable to BNP and NT-proBNP, and therefore omitted from analyses.

[a]Values recorded as % detection [median (interquartile range)].

*$P < 0.05$.

use of baseline and follow-up measurements for association with mortality observed for BNP and BNP 5–32 in the present study may reflect different responses to treatment, with BNP levels being affected by treatment but not BNP 5–32 levels as a result of differential peptide processing in HF patients. BNP molecular forms may provide a more treatment-independent outcome biomarker. In the era of peptidase inhibitors (i.e. sacubitril/valsartan, dipeptidyl peptidase-4 inhibitors), monitoring NPs including molecular forms might allow further insight into NP processing that appear to be altered in HF.

As limitations, BIOSTAT-CHF was a non-randomised observational study, therefore it is not possible to infer causality to our findings or provide a mechanistic explanation. This study involved only European centres, and 99% of patients were Caucasian; therefore, the findings of this study may not be representative of HF patients at a global level.

In conclusion, findings from the BIOSTAT-CHF study, as a real-world cohort, support the role of serial measurement of NPs in clinical practice, with follow-up BNP and BNP

Table 2 Independent prediction abilities of baseline natriuretic peptides for overall outcomes of death and death/heart failure

Multivariate Cox model	Mortality			Mortality/HF		
	HR	95% CI	*P*-value	HR	95% CI	*P*-value
BNP[b]	1.99	1.23–3.23	0.005*	1.72	1.25–2.37	0.001*
BNP 5–32[a]	2.01	1.26–3.20	0.003*	1.54	1.14–2.08	0.005*
NT-proBNP[b]	1.85	1.15–2.99	0.012*	2.33	1.69–3.22	<0.001*

BNP; B-type natriuretic peptide; CI, confidence interval; HF, heart failure; HR, hazard ratio; NT-proBNP, N-terminal pro B-type natriuretic peptide.
The compact risk model for mortality adjusted for age, haemoglobin, blood urea and use of beta-blocker at baseline. The compact risk model for mortality/HF included age, previous HF hospitalisation, peripheral oedema, systolic blood pressure, haemoglobin, sodium and use of beta-blocker at baseline.
[a] Dichotomised according to detection or no detection of the peak.
[b] Values were log transformed.

Table 3 Response to guideline-based treatment for B-type natriuretic peptide (BNP) and BNP 5–32

Dose up-titration		BNP (pg/mL)			BNP 5–32[a]		
	n	V1	V2	*P*-value	V1	V2	*P*-value
ACEi							
<50%	325	228 [100–467]	161 [69–420]	0.359	0.3 [0.0–0.5]	0.0 [0.0–0.3]	<0.001*
≥50%	432	169 [77–344]	114 [39–283]	0.001*	0.0 [0.0–0.5]	0.0 [0.0–0.0]	<0.001*
Beta-blocker							
<50%	424	183 [85–390]	142 [54–382]	0.389	0.2 [0.0–0.5]	0.0 [0.0–0.3]	<0.001*
≥50%	333	208 [88–413]	125 [43–291]	<0.001*	0.0 [0.0–0.5]	0.0 [0.0–0.0]	<0.001*
Both drugs							
Both <50%	684	200 [85–408]	141 [56–382]	0.362	0.2 [0.0–0.5]	0.0 [0.0–0.3]	<0.001*
Both ≥50%	73	206 [86–391]	121 [37–251]	<0.001*	0.0 [0.0–0.5]	0.0 [0.0–0.0]	<0.001*

ACEi, angiotensin-converting enzyme inhibitor; BNP, B-type natriuretic peptide; V1, visit 1 (enrolment); V2, visit 2 (9-month follow-up) <50% less than 50% of optimal recommended dosage, ≥50% of optimal recommended dosage.
Values are reported as median [interquartile range].
[a]BNP 5–32 values reported as a ratio of the mass spectral peak signal intensity against adrenocorticotropic hormone (internal reference standard).

Table 4 Cox models of baseline B-type natriuretic peptide (BNP) 5–32, follow-up BNP 5–32, and combination of BNP 5–32 detection to illustrate whether their combination can better explain all-cause mortality

Serial measurement	Model chi-square	Chi-square if term removed	HR for all-cause mortality (95% CI)	*P*-value
BNP[a] (V1) only	16.0	16.0	2.04 (1.44–2.90)	<0.001*
BNP[a] (V2) only	67.1	67.1	4.03 (2.88–5.65)	<0.001*
BNP[a] (V2) +	66.7	75.1	4.00 (2.71–5.91)	<0.001*
BNP[a] (V1)		0.4	1.04 (0.67–1.60)	0.878
BNP 5-32[b] (V1) only	18.8	18.8	2.14 (1.50–3.04)	<0.001*
BNP 5-32[b] (V2) only	64.3	64.3	3.77 (2.66–5.34)	<0.001*
BNP 5-32[b] (V2) +	69.8	52.2	3.28 (2.28–4.73)	<0.001*
BNP 5-32[b] (V1)		5.5	1.61 (1.09–2.37)	0.017*

BNP, B-type natriuretic peptide; CI, confidence interval; HR, hazard ratio; V1, visit 1; V2, visit 2.
Univariate Cox regression analysis was performed initially for (i) baseline measurement, then for (ii) follow-up measurement, and finally for (iii) baseline + follow-up generating a chi-square for the overall model and also chi-square values for the contribution of the individual variables to the overall model, hence chi-square if term removed.
[a]Values were log transformed.
[b]Dichotomised according to detection or no detection of the peak.

5–32 levels adding value to risk stratification in HF patients. Future studies are needed in cohorts with NP-modulating treatment (i.e. peptidase inhibitors).

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Supplementary Information

Additional supporting information may be found online in the Supporting Information section at the end of the article.

Appendix S1. Supporting Information.

Table S1. (*A*) Independent prediction abilities of baseline natriuretic peptides for overall outcomes of death and death/heart failure with addition of eGFR, blood pressure and past history of diabetes mellitus. (*B*) General linear model for response to guideline-based treatment for BNP and BNP 5–32.

Figure S1. Kaplan–Meier survival curves to show association with outcome of death/heart failure for serial changes in BNP levels in patients that did not achieve a ≥50% dose of guideline treatment up-titration.

Figure S2. Kaplan–Meier survival curves to show association with outcome of death/heart failure for serial detection of BNP 5–32 in patients that did not achieve a ≥50% dose of guideline treatment up-titration.

Figure S3. Kaplan–Meier survival curves to show association with outcome of death/heart failure and death for serial changes in BNP levels following guideline treatment up-titration.

Figure S4. Kaplan–Meier survival curves to show association with outcome of death/heart failure and death for serial detection of BNP 5–32 following guideline treatment up-titration.

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References

1. Ponikowski P, Voors AA, Anker SD, Bueno H, Cleland JG, Coats AJ, Falk V, Gonzalez-Juanatey JR, Harjola VP, Jankowska EA, Jessup M, Linde C, Nihoyannopoulos P, Parissis JT, Pieske B, Riley JP, Rosano GM, Ruilope LM, Ruschitzka F, Rutten FH, van der Meer P. 2016 ESC guidelines for the diagnosis and treatment of acute and chronic heart failure: the Task Force for the diagnosis and treatment of acute and chronic heart failure of the European Society of Cardiology (ESC). Developed with the special contribution of the Heart Failure Association (HFA) of the ESC. *Eur J Heart Fail* 2016;**18**:891–975.
2. Mueller C, McDonald K, de Boer RA, Maisel A, Cleland JG, Kozhuharov N, Coats AJ, Metra M, Mebazaa A, Ruschitzka F, Lainscak M, Filippatos G, Seferovic PM, Meijers WC, Bayes-Genis A, Mueller T, Richards M, Januzzi JL Jr; Heart Failure Association of the European Society of Cardiology. Heart Failure Association of the European Society of Cardiology practical guidance on the use of natriuretic peptide concentrations. *Eur J Heart Fail* 2019;**21**:715–731.
3. Januzzi JL, Troughton R. Are serial BNP measurements useful in heart failure management? Serial natriuretic peptide measurements are useful in heart failure management. *Circulation* 2013;**127**:500–507 discussion 508.
4. Latini R, Masson S, Wong M, Barlera S, Carretta E, Staszewsky L, Vago T, Maggioni AP, Anand IS, Tan LB. Incremental prognostic value of changes in B-type natriuretic peptide in heart failure. *Am J Med* 2006;**119**:70.e23–30.
5. Desai AS. Are serial BNP measurements useful in heart failure management? Serial natriuretic peptide measurements are not useful in heart failure management: the art of medicine remains long. *Circulation* 2013;**127**:509–516; discussion 516.
6. Suzuki T, Israr MZ, Heaney LM, Takaoka M, Squire IB, Ng LL. Prognostic role of molecular forms of B-type natriuretic peptide in acute heart failure. *Clin Chem* 2017;**63**:880–886.
7. Suzuki T, Yazaki Y, Voors AA, Jones DJ, Chan DC, Anker SD, Cleland JG, Dickstein K, Filippatos G, Hillege HL. Association with outcomes and response to treatment of trimethylamine N-oxide in heart failure: results from BIOSTAT-CHF. *Eur J Heart Fail* 2019;**21**:877–886.
8. Felker GM, Anstrom KJ, Adams KF, Ezekowitz JA, Fiuzat M, Houston-Miller N, Januzzi JL, Mark DB, Piña IL, Passmore G, Whellan DJ, Yang H, Cooper LS, Leifer ES, Desvigne-Nickens P, O'Connor CM. Effect of natriuretic peptide-guided therapy on hospitalization or cardiovascular mortality in high-risk patients with heart failure and reduced ejection fraction: a randomized clinical trial. *JAMA* 2017;**318**:713–720.
9. Januzzi JL, Ahmad T, Mulder H, Coles A, Anstrom KJ, Adams KF, Ezekowitz JA, Fiuzat M, Houston-Miller N, Mark DB, Piña IL, Passmore G, Whellan DJ, Cooper LS, Leifer ES, Desvigne-Nickens P, Felker GM, O'Connor CM. Natriuretic peptide response and outcomes in chronic heart failure with reduced ejection fraction. *J Am Coll Cardiol* 2019;**74**:1205–1217.
10. Kubánek M, Goode KM, Lánská V, Clark AL, Cleland JG. The prognostic value of repeated measurement of N-terminal pro-B-type natriuretic peptide in patients with chronic heart failure due to left ventricular systolic dysfunction. *Eur J Heart Fail* 2009;**11**:367–377.
11. Zile MR, Claggett BL, Prescott MF, McMurray JJ, Packer M, Rouleau JL, Swedberg K, Desai AS, Gong J, Shi VC, Solomon SD. Prognostic implications of changes in N-terminal pro-B-type natriuretic peptide in patients with heart failure. *J Am Coll Cardiol* 2016;**68**:2425–2436.