B-type natriuretic peptide, a marker of asymptomatic left ventricular dysfunction in type 2 diabetic patients

J.-P. Albertini\textsuperscript{a,}* , R. Cohen\textsuperscript{b}, P. Valensi\textsuperscript{c}, R.N. Sachs\textsuperscript{d}, J.-C. Charniot\textsuperscript{d}

\textsuperscript{a} Laboratory of Biochemistry, Avicenne Hospital, AP–HP, 125, route de Stalingrad, 93009 Bobigny cedex, France
\textsuperscript{b} Department of Internal Medicine and Endocrinology, Avicenne Hospital, AP–HP, Bobigny, France
\textsuperscript{c} Department of Endocrinology Diabetology Nutrition, Jean-Verdier Hospital, Paris Nord University, AP–HP, CRNH-Ile-de-France, Bondy, France
\textsuperscript{d} Department of Cardiology, Avicenne Hospital, AP–HP, Bobigny, France

Received 3 May 2007; received in revised form 13 February 2008; accepted 24 February 2008
Available online 2 July 2008

Abstract

Aim. – To evaluate BNP in assessing LV functions in asymptomatic type 2 diabetic patients.

Methods. – BNP was measured in 91 consecutive patients with type 2 diabetes mellitus. According to Doppler echocardiography, patients were first separated into three categories: normal LV function, or isolated diastolic or systolic LV dysfunction. As some patients with diastolic dysfunction were treated for hypertension, the population was divided into four groups: groups 1, 2 and 3 all had no antihypertensive treatment, and had normal LV function, and isolated diastolic and systolic LV dysfunction, respectively; and group 4 were being treated with antihypertensive drugs and had diastolic LV dysfunction.

Results. – In group 1, BNP levels (13 \(\pm\) 2 ng/L) were lower than in group 2 (87 \(\pm\) 20 ng/L, \(P < 0.0001\)) or group 3 (213 \(\pm\) 32 ng/L, \(P < 0.0001\)), but were similar to those of group 4 (32 \(\pm\) 6 ng/L, \(P = 0.14\)). ROC analysis revealed a rule-out value of 23 ng/L for group 1 versus group 2, and of 239 ng/L for group 2 versus group 3. In groups 1, 2 and 3 taken together, BNP levels were correlated with urinary albumin excretion rate (\(r = 0.80\), \(P < 0.0001\)) and pulse pressure (\(r = 0.65\), \(P < 0.0001\)). In group 4, patients receiving ACE inhibitors had lower BNP levels than those receiving \(\beta\)-blockers.

Conclusion. – BNP can be used to pre-screen asymptomatic type 2 diabetic patients with LV dysfunction, and may reveal vascular remodelling in type 2 diabetes mellitus.

© 2008 Elsevier Masson SAS. All rights reserved.

Résumé

Peptide natriurétique de type B, marqueur d’une dysfonction ventriculaire gauche asymptomatique chez des diabétiques de type 2.

Objectif. – Doser le peptide natriurétique de type B (BNP) plasmatique chez des diabétiques de type 2 asymptomatiques pour déceler une dysfonction précoce du ventricule gauche (VG).

Méthodes. – Les concentrations plasmatiques de BNP ont été mesurées chez 91 diabétiques de type 2. Selon les résultats de l’écho-Doppler, les patients ont d’abord été répartis en trois groupes : fonction VG normale, ou dysfonction diastolique ou systolique isolée. Certains patients présentant une fonction diastolique altérée étant traités pour hypertension, la répartition a été modifiée en quatre groupes : groupes 1, 2 et 3 sans traitement antihypertenseur et remplissant les trois critères, fonction VG normale, ou dysfonction diastolique ou systolique isolée, et groupe 4 avec une dysfonction diastolique et traité par antihypertenseurs.

© 2018 Elsevier Masson SAS. Tous droits réservés. - Document téléchargé le 10/12/2018 Il est interdit et illégal de diffuser ce document.

Abbreviations: ACE, angiotensin-converting enzyme; AD, aorta diameter; ARAII, angiotensin-II receptor antagonists; AUC, area under the curve; BNP, B-type natriuretic peptide; DTE, deceleration time of E-wave; LA, left atrium; LV, left ventricular; LVEDD, LV end-diastolic diameter; LVEF, LV ejection fraction; LVESD, LV end-systolic diameter; LVFS, LV fractional shortening; LVM, LV mass index; PP, pulse pressure; ROC, receiver operating characteristic; UAER, urinary albumin excretion rate.

* Corresponding author.

E-mail address: jean-paul.albertini@avc.aphp.fr (J.-P. Albertini).

doi:10.1016/j.diabet.2008.02.004
1. Introduction

The role of diabetes mellitus in congestive heart failure was recognized for the first time in early results from the Framingham cohort [1]. As shown in the Studies of Left Ventricular Dysfunction (SOLVD) [2] and registry, subclinical LV dysfunction is a frequent condition in asymptomatic patients with diabetes mellitus, and cardiovascular disease (CVD) is the underlying cause of death in more than 60% of type 2 diabetic patients [3].

Early diagnosis and treatment of cardiac dysfunction may lead to improved survival in diabetic patients. Identification at the asymptomatic stage is mainly based on echocardiography. This method assesses abnormal LVEF and systolic dysfunction, and can also evaluate diastolic filling abnormalities using transmitral Doppler flow velocity. In diabetic cardiomyopathy, abnormalities of diastolic function often go clinically unrecognized and precede systolic dysfunction [4]. However, in diabetes, worsening of diastolic function remains controversial [5]. Also, echocardiography is of limited availability and is relatively expensive [6]. A simple, easily available and non-invasive biochemical testing method would be ideal for screening for subclinical cardiac dysfunction, and could help to improve clinical outcomes in diabetic patients.

BNP is secreted by ventricular cardiomyocytes and released into the bloodstream in response to haemodynamic stress mainly induced by pressure overload, leading to hypertrophy and, thus, cardiac dysfunction [7]. BNP has a vasorelaxant effect on systemic circulation, and an inhibitory effect on renin–angiotensin–aldosterone secretion [8]. Plasma BNP levels are useful in the diagnosis and prognosis of LV dysfunction [9]. They are influenced by several factors, such as age [10] and renal failure, both of which raise BNP, and obesity, which reduces BNP [11]. In acute dyspnoea, a clear increase in BNP may point to congestive heart failure. Studies have suggested that BNP may also be effective in screening for early and asymptomatic LV dysfunction with intact systolic function, and can even rule out heart failure, as levels below 100 ng/L have a high negative predictive value [12]. In addition, two studies of diabetic patients have suggested that levels of plasma N-terminal pro-BNP may be predictive of cardiovascular mortality in type 1 diabetics with nephropathy [13] and of cardiovascular events in type 2 diabetics with microalbuminuria [14]. The relationship between plasma BNP levels and microalbuminuria has so far been examined in only a few studies, with contradictory results [14–16]. The aim of the present study is to evaluate the predictive value of plasma BNP levels for the early detection of impaired LV function in consecutive asymptomatic hospitalized type 2 diabetic patients.

2. Subjects and methods

One hundred and twenty-one consecutive asymptomatic patients with type 2 diabetes mellitus and no cardiac history were prospectively recruited after being admitted to the Avicenne Hospital endocrinology unit. The majority of these patients had poorly controlled type 2 diabetes (Table 1). Thirty patients were excluded from the study due to a previous or suspected history of heart disease, intrinsic lung or overt renal disease, incomplete echocardiographic data or poor echogenicity. From the clinical data analysis of the 91 patients enrolled (47 men, 44 women; mean age ± S.D.: 60 ± 14, 61 ± 15 years, respectively), 42 were further found to be treated with antihypertensive drugs, including β-blockers, ACE inhibitors (ACEI), ARALI, diuretics and calcium-channel inhibitors either alone or in combination, for at least six months before entry into the study. All clinical, biological and treatment data were collected upon admission. Hypertension was defined as a blood pressure more than 140/90 mmHg, according to American Diabetes Association (ADA) recommendations [17]. PP, a component of blood pressure that increases when arterial compliance is reduced, was calculated as the difference between systolic and diastolic blood pressures. The study received the approval of our institutional board, and written informed consent was obtained from all the participants.

3. Analytical methods

All blood samples were collected at baseline after an overnight fast.

3.1. B-type natriuretic peptide

Venous blood samples for BNP measurement were collected by venipuncture into an EDTA tube. Within one hour of the draw time, whole-blood BNP concentrations were measured by a Triage® BNP meter, using a point-of-care fluorescence immunoassay (Biosite Diagnostics, France). Assays were com-
Echocardiographic data

Current treatment (%)

Plasma glucose levels were measured by the glucose oxidase method (Bayer Diagnostics, Puteaux, France). HbA1c levels (normal range 4–5.5%) were measured by ion-exchange high-performance liquid chromatography (HPLC), normalized in accordance with the Diabetes Control and Complications Trial (DCCT) reference method, but improved with a CV less than 4% (Bio-Rad, Hercules, CA, USA). Serum lipid levels (triglycerides, total cholesterol, HDL-cholesterol and LDL-cholesterol) were measured by automated enzymatic methods (Randox, Crumlin, UK). Serum C-peptide levels were determined using a chemiluminescence assay (ImmunoLite C-peptide, DPC, Los Angeles, CA, USA). Serum creatinine was measured by automated Jaffé method (Bayer Diagnostics) and UAER by a polyethylene glycol-enhanced immunoturbidimetric assay (Bayer Diagnostics). Microalbuminuria was defined as a UAER equal to 30–300 mg/24 h.

3.2. Biochemical parameters

Plasma glucose levels were measured by the glucose oxidase method (Bayer Diagnostics, Puteaux, France). HbA1c levels (normal range 4–5.5%) were measured by ion-exchange high-performance liquid chromatography (HPLC), normalized in accordance with the Diabetes Control and Complications Trial (DCCT) reference method, but improved with a CV less than 4% (Bio-Rad, Hercules, CA, USA). Serum lipid levels (triglycerides, total cholesterol, HDL-cholesterol and LDL-cholesterol) were measured by automated enzymatic methods (Randox, Crumlin, UK). Serum C-peptide levels were determined using a chemiluminescence assay (ImmunoLite C-peptide, DPC, Los Angeles, CA, USA). Serum creatinine was measured by automated Jaffé method (Bayer Diagnostics) and UAER by a polyethylene glycol-enhanced immunoturbidimetric assay (Bayer Diagnostics). Microalbuminuria was defined as a UAER equal to 30–300 mg/24 h.

3.3. Doppler echocardiography

Upon admission, patients were subjected to two-dimensional, M-mode spectral and colour flow Doppler transthoracic echocardiography, using an ATL 5000 HDI machine equipped with a 2.5 MHz probe and TDI (tissue Doppler imaging). Two-dimensional imaging analyses were performed in the standard fashion with parasternal long- and short-axis views, and apical four- and two-chamber views. Measurements were performed by one cardiologist (J.-C. C.), who was unaware of the patients’ clinical condition or treatment, or BNP levels.
Normal ventricular function was defined as normal LVEDD (35–55 mm) and LVESD (25–36 mm), and LVFS >30%. LVEF was estimated using the Simpson method and was considered normal when greater or equal to 50%. Pulsed Doppler analysis of mitral inflow included measurements of mitral valve early peak filling velocity (E), late peak filling velocity (A), E/A ratio and DTE. Pulsed wave TDI of the mitral annulus was obtained from the apical four-chamber view. A 1.7 cm² sample volume was placed on the lateral side of the mitral annulus to obtain early (Ea) peak velocities. The E/Ea index was calculated as proposed by Nagueh et al. [18]. Diastolic dysfunction was defined as a normal or mildly impaired LVEF (>50%) and as:

- an ‘impaired relaxation’ pattern with an E/A ratio less than one, or greater than one if associated with an E/Ea index greater than ten and early reduction of diastolic velocity at the lateral annulus of less than 8 cm/s;
- a ‘restrictive’ pattern with an E/A ratio greater than two, or between one and two with a DTE less or equal to 130 ms;
- a ‘pseudo-normal’ pattern with an E/A ratio of 1–2 with a DTE of 150–220 ms.

The LVMI (ratio of LV mass/body surface area) was considered normal if less than 110 g/m² in women and less than 130 g/m² in men [4]. Systolic LV dysfunction was defined as an LVEF less than 45% and/or LVFS less than 30%.

4. Statistical methods

Statistical analyses were performed using SPSS v10 (SPSS France) and JMP v5 (SAS Institute, Cary, NC, USA) software. Results are summarized as means ± S.D. Normality for each variable was tested using Fisher’s y² parameter and, because of the skewed distributions of the analyzed data, especially the BNP levels and UAER, the comparisons used non-parametric statistical procedures or parametric tests using natural logarithmic transformation. Multivariate regressions used backward/forward likelihood-ratio procedures, a P value ≤ 0.05 for inclusion and a P value < 0.10 for removal. Correction for multiple comparisons was not performed in the present study. The diagnostic accuracy of BNP alone was compared with echocardiographic and Doppler data, and statements used nominal logistic regressions completed by ROC curves. Results are expressed with the area under the ROC curve (AUC) as an index of the overall test for accuracy. All calculations were performed using a significance level of 0.05.

5. Results

Based on Doppler echocardiographic findings, patients were first divided into three categories: normal LV function; isolated diastolic LV dysfunction; and isolated systolic LV dysfunction. To explain discrepancies in BNP levels compared with echocardiographic data, we reconsidered the clinical data and found that some type 2 diabetic patients with diastolic dysfunction were receiving antihypertensive treatments that affected BNP levels. Therefore, we subdivided these patients into two further groups.

Finally, and according to BNP values, patients were divided into four groups (Table 1): group 1 (n = 21), 2 (n = 18) and 3 (n = 10) had all received no antihypertensive treatment, and had normal LV function, and isolated diastolic and systolic LV dysfunction, respectively, while group 4 had been treated with antihypertensive drugs and had diastolic LV dysfunction—either a filling abnormality (34 patients) or pseudo-normalization (8 patients).

5.1. Patients not receiving antihypertensive treatment (groups 1–3)

5.1.1. Clinical and biological parameters

Patients with either diastolic (group 2) or systolic dysfunction (group 3) were significantly older than patients with normal LV function (group 1) (P < 0.0001). Among these three groups, no significant differences were observed in terms of gender (P = 0.97), HbA1c (P = 0.4), serum creatinine (P = 0.33) or lipid parameters (P > 0.07) (Table 1).

5.1.2. Echocardiographic and Doppler data

LA, LA/AD ratio, LVMI, LVEDD and DTE did not differ across the three groups (P > 0.2), whereas LVESD, LVEF, LVFS, and E/A ratio and E/Ea index were significantly different (P = 0.01, P < 0.0001, P = 0.0001 and P < 0.0001, respectively). E/A and E/Ea were both significantly lower in group 2 compared with group 1 (P < 0.0001). LVMI was higher in groups 2 and 3 compared with group 1 (P < 0.001).

5.1.3. BNP levels

BNP levels were significantly higher in groups 2 and 3 compared with group 1 (87 ± 20 ng/L and 213 ± 32 ng/L versus 13 ± 2 ng/L, respectively; P = 0.0001) (Fig. 1A). In groups 1, 2 and 3 taken together, BNP levels correlated with age (r = 0.51, P < 0.0001), but not with body mass index (BMI) (r = −0.30, P = 0.40). Interestingly, when only groups 1 and 2 were taken together, BNP levels correlated with age (r = 0.45, P = 0.01), UAER (r = 0.80, P < 0.0001) (Fig. 2A), PP (r = 0.65, P < 0.0001) (Fig. 2B) and LVMI (r = 0.77, P < 0.0001) (Fig. 2C), but not with BMI, and diastolic and systolic blood pressures.

After controlling for age in the three groups taken together, BNP levels partially correlated with UAER, PP, LVFS and DTE (r = 0.74, P < 0.0001; r = 0.37, P = 0.02; r = −0.35, P = 0.03; and r = 0.4, P = 0.01, respectively) whereas taking only groups 1 and 2, BNP was partially correlated with UAER, PP, LVMI and the E/A ratio (r = −0.80, P < 0.0001; r = 0.65, P < 0.0001; r = −0.76, P < 0.0001; and r = 0.35, P = 0.03, respectively).

Multivariate analysis using both forward and backward methods showed a significant and independent positive correlation between BNP levels and UAER and PP, and a negative correlation between BNP levels and LVEF (multiple r = 0.79, P < 0.0001; and for each independent variable: P < 0.001, P < 0.03 and P < 0.03, respectively). The same analyses using only groups 1 and 2 resulted in a positive correlation between BNP levels and UAER, PP and LVMI, but a negative correlation with LVEF (multiple r = 0.95, P < 0.0001; and for each independent variable: P = 0.01, P = 0.01, P = 0.02 and P = 0.02, respectively).
Fig. 1. A: Box plot of the four patient groups according to their echocardiographic data, BNP levels and use or not of antihypertensive treatment. Boxes include the medians, interquartile ranges and outliers, and means are indicated by crosses. The plot is divided into two parts: the asymptomatic type 2 diabetics who did not receive antihypertensive drugs are on the left, and the overall BNP levels in type 2 diabetics treated for overt hypertension are on the right; B: receiver operating characteristic (ROC) curves for BNP accuracy in group 2 versus group 1 and in group 3 versus group 2.

Fig. 2. For groups 1 and 2 taken together, correlations between natural logarithm-transformed BNP (Ln BNP) and: A: UAER, \( r = 0.80, P < 0.0001 \); B: pulse pressure, \( r = 0.65, P < 0.0001 \); C: LVMI, \( r = 0.77, P < 0.0001 \).
5.2. Patients treated for overt hypertension (group 4)

In these 42 type 2 diabetic patients (Fig. 1), echocardiographic and Doppler data favoured LV diastolic dysfunction. However, BNP levels fell dramatically (32 ± 6 ng/L) in comparison to group 2 (87 ± 20 ng/L) (P = 0.003), with no significant difference compared with those of group 1, who had normal BNP levels (13 ± 2 ng/L, P = 0.13). LVFS and E/A were higher, and DTE lower, in group 4 than in group 2. Furthermore, the highest BNP levels (46 ± 13 ng/L) were found in patients treated with β-blockers, and the lowest (27 ± 8 ng/L) in those treated with ACEI or ARAII.

6. Discussion

In the present study, BNP levels in clinically asymptomatic type 2 diabetic patients were able to detect LV dysfunction, whether systolic or diastolic, and possibly its normalization through cardiac treatments, as well as the influence of endothelial dysfunction associated with microalbuminuria.

Patients were mainly referred to our department to assess diabetes complications and to improve glycaemic control. Although blood glucose was poorly controlled, BNP concentrations are known to be unaffected by elevated glucose levels [19].

Many studies have confirmed the relevance of BNP assays compared with echocardiographic data, but few have looked at clinically asymptomatic type 2 diabetics. Our study categorized type 2 diabetic patients into four well-defined groups according to their echocardiographic findings: normal LV function (group 1); diastolic LV dysfunction (group 2); systolic LV dysfunction (group 3); and patients treated for hypertension (group 4).

In group 1, BNP levels were normal and similar to those found in the general, asymptomatic population [20]. In patients with diastolic LV dysfunction (group 2), BNP levels were considered abnormal (87 ± 20 ng/L) as the prognosis is worsened when BNP levels are below the threshold of 100 ng/L, which has a strong negative predictive value in an emergency, but is associated with an increased risk of heart failure, atrial fibrillation or stroke [20]. In group 3, BNP levels were markedly increased in patients with asymptomatic systolic dysfunction and were similar to those previously reported by Epshteyn et al. [21].

In patients who had not received antihypertensive treatment, there were significant correlations between BNP levels and:

- UAER, which might indicate endothelial dysfunction, setting up a prognostic factor for subsequent cardiovascular events in diabetic patients [22];
- LVMI, an indication of LV remodelling;
- PP, which accounts for the vascular remodelling [23];
- but not with creatininemia.

Although correlations with UAER need to be treated with caution as UAER was measured only once, the present data suggest that BNP might be an early in vitro marker for diffuse endothelial dysfunction, which has been shown to induce atherothrombotic changes [24] and to contribute to future cardiovascular complications, as confirmed by Framingham observations. Indeed, correlations have been reported between UAER and brachial hyperaemic responses to ischaemia [25] and coronary endothelial dysfunction [26]. Two recent studies have also suggested that natriuretic peptides may provide prognostic cardiovascular evaluation in addition to microalbuminuria in patients with either type 1 or 2 diabetes [13,14]. In addition, Chong et al. showed a significant inverse correlation between flow-mediated endothelial-dependent vasodilation and BNP levels [27].

Among the pitfalls of BNP, it has been shown that antihypertensive treatments have an effect upon LV function. Another interesting feature of our study was seen in group 4, where the heterogeneity of antihypertensive treatments was found to perturb BNP levels which, overall, had a mean level similar to that of group 1. Considerable evidence shows that β-blockers, ACEI and ARAII improve LV filling while reducing LV hypertrophy as well as PP. With ARAII treatment, Okin et al. [28] and Devereux et al. [29] reported a greater regression of LV hypertrophy than with β-blockers. Among our 42 treated patients, 34 had a persistent filling impairment and eight showed pseudo-normalization—not mentioned in previous echocardiographic studies. Furthermore, in patients treated with either ACEI or ARAII, BNP levels tended to be lower than in patients treated with β-blockers. In line with this, losartan has been shown to reduce BNP levels, and atenolol to increase BNP, in hypertensive patients with LV hypertrophy [30].

To rule out incipient silent myocardial ischaemia, non-invasive tests such as stress-test electrocardiography or dipyridamole–thallium scintigraphy should have been performed, as well as TDI of mitral annular motion and pulmonary venous flow, both of which are now recognized as giving, albeit indirectly, better estimates of LV filling pressure. Only the early peak diastolic (Ea) was obtained from routine echocardiography. Unlike the study by Cosson et al. [31], we evaluated BNP levels and correlated them with standard Doppler echocardiography. We did not evaluate the new Doppler indices for diastolic dysfunction, but only the usual parameters, as such recent evaluations were not planned for in our protocol, which used only the routine evaluations. Nevertheless, using conventional Doppler echocardiography in asymptomatic, older type 2 diabetes patients who were free of structural heart disorders,
Valle et al. [32] reported high prevalence rates of subclinical LV diastolic dysfunction (39/76 patients) with intact systolic function, which accords with our results. Also, in the Valle study, the patients’ ages were similar to ours (61 ± 7 versus 60 ± 2 years).

Finally, the small size of our sample and survey did not allow any inference of useful indices to predict cardiovascular events that might arise after screening.

It is noteworthy that senior patients with type 2 diabetes are rarely untreated for hypertension, which might explain the relatively small numbers included in groups 1, 2 and 3. Further studies in vivo are probably needed to clarify whether or not endothelial dysfunction is more severe in asymptomatic diabetic patients who have both microalbuminuria and slightly elevated BNP levels.

7. Conclusion

Our study shows significant correlations between BNP levels and microalbuminuria, PP and LVMI. Taken altogether, these markers may, in vitro, indicate a diffuse endothelial dysfunction and probably the prognosis as well. Despite the relative instability of BNP, its measurement is reproducible, and easily and rapidly obtained at a low cost. There was no overlap between BNP levels in patients with normal LV function and those with diastolic or systolic LV dysfunction. BNP levels could also be used for repeat evaluations of an occult LV dysfunction in patients who are periodically assessed for diabetes complications. In this way, BNP could provide the deciding factor for referring secondary diabetic patients for echocardiography. However, as antihypertensive treatments can modify BNP levels, this affect needs to be evaluated through controlled studies specifically in asymptomatic patients with diabetes.

Acknowledgments

The authors are grateful to Mr Fabrice Heurtin for his invaluable help in writing this manuscript. The authors also wish to thank Mrs Jocelyne Valladeau and Mrs Dina Dehaupas for their technical help.

References

[27] Chong AY, Blann AD, Patel J, Freestone B, Hughes E, Lip GY. Endothelial dysfunction and damage in congestive heart failure: relation of flow-mediated dilation to circulating endothelial cells, plasma


