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N-terminal-pro-brain natriuretic peptide, but not brain natriuretic peptide, is increased in patients with severe obesity

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Abstract

Elevated body mass index (BMI) has been reported as a risk factor for heart failure. Prevention of heart failure through identification and management of risk factors and preclinical phases of the disease is a priority. Levels of natriuretic peptides as well as activity of their receptors have been found altered in obese persons with some conflicting results. We investigated cardiac involvement in severely obese patients by determining N-terminal-pro-brain natriuretic peptide (NT-proBNP) and brain natriuretic peptide (BNP) and attempting to correlate the levels of these peptides in serum and plasma, respectively, with BMI, duration of obesity, waist circumference, and echocardiographic parameters. Thirty-three patients with severe obesity (mean BMI: 46.39 kg/m^2, mean age: 39 years) were studied. The control group contained 30 healthy age-matched individuals (BMI: <25 kg/m^2, mean age: 43 years). The *t*-test and Spearman correlation were used for statistical analysis. Log-NT-proBNP was significantly higher (P = 0.003) in obese patients (mean 1.67, 95% CI: 1.50-1.83 log pg/mL) compared to controls (mean: 1.32, 95% CI: 1.17-1.47 log pg/mL). The Log-NT-proBNP concentration correlated with duration of obesity (r = 0.339, P < 0.004). No difference was detected in the Log-BNP concentration (P = 0.63) of obese patients (mean: 0.73, 95% CI: 0.46-1.00 log pg/mL) compared to controls (mean: 0.66, 95% CI: 0.51-0.81 log pg/mL). NT-proBNP, but not BNP, is increased in severely obese patients and its concentration in serum is correlated with duration of obesity. NT-proBNP may be useful as an early diagnostic tool for the detection of cardiac burden due to severe obesity.

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Introduction

Severe obesity is now recognized as a risk factor for heart failure (1). Epidemiological (2), echocardiographic (3), and autopsy (4,5) studies have identified obesity cardiomyopathy as an isolated clinical entity. In these studies, elevated body mass index (BMI) is described as a risk factor for left ventricular remodeling and overt heart

failure.

Left ventricular enlargement and eccentric hypertrophy are the most common morphological cardiac abnormalities in obese individuals (3). This cardiac remodeling depends on the severity and duration of obesity and the influence of adverse loading conditions (6,7).

N-terminal-pro-brain natriuretic peptide (NT-proBNP) and brain natriuretic peptide (BNP) are useful for the diagnosis of heart failure, and their high levels in serum and plasma, respectively, are related to wall stress, which is often increased in severe obesity. They are sensitive markers of cardiac dysfunction and may be useful as early diagnostic tools for the detection of cardiac overload in this group of patients (8,9). However, NT-proBNP has a slower plasma clearance than when compared to the biologically active peptide BNP, resulting in higher circulating concentrations of NT-proBNP, although both peptides are released by cardiomyocytes on an equimolar basis (10). Furthermore, the different mechanisms of plasma clearance (neutral endopeptidase clearance receptors for BNP versus renal clearance for NT-proBNP) may be different in obese patients. The potential differences of either BNP or NT-proBNP concentration in plasma and serum, respectively, for the diagnosis of cardiac involvement in patients with severe obesity have not been established.

The aim of the present study was to compare BNP and NT-proBNP with respect to their diagnostic utility for the detection of cardiac involvement in patients with severe obesity and to correlate their levels in plasma, BMI, duration of obesity, waist circumference (WC), and echocardiographic parameters.

Material and Methods

Patients

We selected 33 consecutive patients with level III obesity and BMI >40, 23 females (mean age: 39 years). BMI was defined as weight in kilograms divided by height in square meters (kg/m^2). The exclusion criteria were myocardial infarction (evaluated by Q waves on an electrocardiogram, segmental dysfunction by 2-D echocardiography) and valvar disease (evaluated by clinical examination and by Doppler echocardiography). Hypertension was defined as systolic blood pressure >140 mmHg or diastolic blood pressure >90 mmHg. A control group comprised 30 healthy, thin individuals (BMI <25, 9 females, mean age: 43 years).

This investigation conforms to the principles outlined in the Declaration of Helsinki. All patients signed a written informed consent form, and the Ethics Committee of the Heart Institute of the University of São Paulo approved the study.

Procedures

Two-dimensional echocardiography complemented by M-mode and color-Doppler recordings was obtained according to the recommendations of the American Society of Echocardiography (11,12). The following indices were obtained: septum and posterior wall left ventricular (LV) thickness in diastole, LV internal end-diastolic dimension, LV internal end-systolic dimension, left atrium size, LV mass, and LV mass index. Also, the ejection fraction was calculated using the Teichholz method. E and A mitral wave peak velocities and E/A ratio were also analyzed.

Serum NT-proBNP levels were measured by immunoassay with detection by electro-chemiluminescence (Roche Diagnostics, São Paulo, SP, Brazil) using 20 µL of serum and polyclonal antibodies that detect epitopes in the N-terminal region (amino acids 1-76) of the proBNP (108 amino acids). Cross-reactivity with other natriuretic peptides (BNP, proANP1, CNP2) and the renin-angiotensin system were <0.001% (data from

the manufacturer). The assay is fully automated using the Elecsys 2010 automated analyzer (Roche Diagnostics).

Plasma BNP levels were measured by a two-site dual-monoclonal immunochemiluminescent assay (Bayer ADVIA Centaur, Bayer Diagnostics, São Paulo, SP, Brazil) using 100 µL of plasma and monoclonal antibodies that detect epitopes in the C-terminal region of the BNP (amino acids 14-21). Both antibodies were supplied by Shionogi Co., Ltd. (Osaka, Japan). The assay was tested for several BNP fragments and other neuropeptides: α-atrial natriuretic peptide; NT-proBNP amino acids 1-21; NT-proBNP 22-46; NT-proBNP 1-46; NT-proBNP 1-76; NT-proBNP 47-76; urodilatin; C-, D-, and V-natriuretic peptides; adrenomedullin-52; angiotensin III; arg-vasopressin (all at 1.0 ng/L). There was no significant cross-reactivity of the ADVIA Centaur assay against other BNP fragments and neuropeptides (cross-reactivities ranging from -2.2% for adrenomedullin to 1.6% for atrial natriuretic peptide).

Blood was collected by venipuncture into plastic evacuated tubes containing EDTA for BNP determination. Samples were kept at room temperature for a maximum of 1 h before centrifugation and plasma separation. Plasma was immediately frozen and stored at -20ºC. The sample was thawed just before the analysis, which was carried out within 6 months of blood collection. BNP is stable for at least 1 year when frozen at -20ºC.

For the determination of NT-proBNP, blood was collected into glass tubes without additives and centrifuged within 1 h. In the meantime, samples were kept at room temperature. The serum was immediately frozen and stored at -20ºC. The sample was thawed just before the analysis, which was carried out within 6 months of blood collection. NT-proBNP is stable for at least 1 year when frozen at -20ºC.

For the BNP assay the coefficient of variation was 4.7% (mean concentration 29.4 pg/mL) and the inter- and intra-assay variations were 4.3 and 1.9%. The upper and lower limit concentrations are 5 and 5000 pg/mL, respectively (data from the manufacturer).

For the NT-proBNP assay the coefficient of variation was 7.0% (mean concentration 57 pg/mL) and the inter- and intra-assay variations were 4.0 and 2.6%, respectively. Concentration ranged from 5 up to 35,000 pg/mL (data from the manufacturer).

The threshold (lower limit of detection) was 5 pg/mL for both assays.

Statistical analysis

NT-proBNP and BNP data were log transformed in order to obtain a normal distribution of the sample that would permit to use parametric methods for statistical analysis. The Student *t*-test for unpaired data was used to compare the log transformed means of the circulating level of the hormone in morbidly obese and control subjects. Spearman correlations were used to assess the relation between hormone levels and BMI, duration of obesity, or WC in morbidly obese subjects. $P < 0.05$ was considered to be statistically significant.

Results

All patients had level III obesity with very high BMI (mean: 46.39 ± 5.02 kg/m^2), long-term duration of obesity ranging from 2 to 40 years (mean: 15.64 ± 10.18 years), WC ranging from 103 to 157 cm (mean: 131.37 ± 14.2 cm). Twenty patients in the obese group had mild hypertension (Table 1).

Log-NT-proBNP ($P = 0.003$) was higher in patients with severe obesity (mean: 1.67, 95% CI: 1.50-1.83 log pg/mL) compared to the control group (mean: 1.32, 95% CI: 1.17-1.47 log pg/mL). Log-NT-proBNP correlated only with duration of obesity in these patients ($P < 0.004$, $r = 0.339$; Figure 1), but

not with WC, BMI (Table 2), echocardiographic parameters (Table 1) and with the presence of hypertension.

However, no differences were found in the Log-BNP (P = 0.63) in patients with severe obesity (mean 0.73, 95% CI: 0.46-1.00 log pg/mL) compared with that in the control group (mean 0.66, 95% CI: 0.51-0.81 log pg/mL).

Table 1. Characteristics of the obese and control groups.

	Obese group	Control group
Age (years)	39 ± 11.2	43 ± 4.8
Sex (female/male)	23/10	9/21
BMI (kg/m^2)	46.39 ± 5.02	23.1 ± 1.8
Duration of obesity (years)	15.64 ± 10.18	
Waist circumference (cm)	131.37 ± 14.2	
Hypertension (patients)	20	
Septum thickness (cm)	1.0 ± 0.15	0.8 ± 0.2
Left ventricular diastolic diameter (cm)	5.13 ± 0.54	4.6 ± 0.6
Left ventricular systolic diameter (cm)	3.39 ± 0.43	2.9 ± 0.5
Left atrial diameter (cm)	3.74 ± 0.42	3.4 ± 0.5
Left ventricular mass (g)	205.31 ± 64.08	
Ejection fraction (%)	62.83 ± 4.76	72 ± 3
Left ventricular mass index (g/m^2)	89.68 ± 21.60	
Log-BNP (pg/mL)	0.73 (CI = 0.46-1.00)	0.66 (CI = 0.51-0.81)
Log-NT-proBNP (pg/mL)	1.67 (CI = 1.50-1.83)	1.32 (CI = 1.17-1.47)

Data are reported as means ± SD for 33 patients and 30 controls, except for Log-BNP and Log-NT-proBNP, which are reported as mean and 95% confidence interval (CI). BNP = brain natriuretic peptide; NT-proBNP = N-terminal-pro-brain natriuretic peptide.

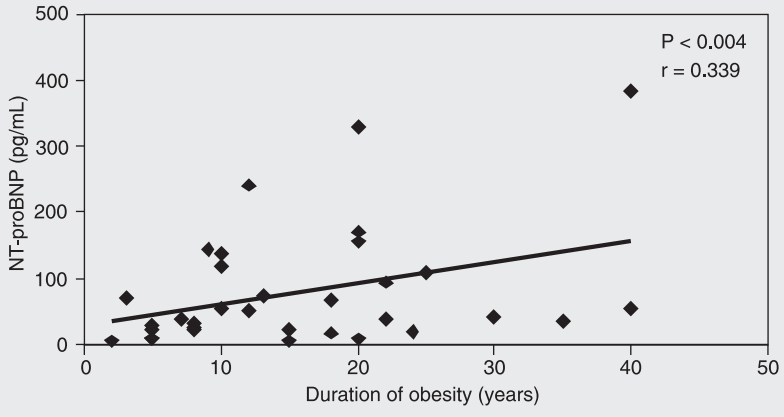

Figure 1. Spearman correlation between Log-N-terminal-pro-brain natriuretic peptide (NT-proBNP) and the duration of obesity (N = 33).

Discussion

The main finding in the present study was that patients with level III obesity had higher NT-proBNP compared to control. Therefore, NT-proBNP could be used as an early marker of cardiac involvement.

Obesity produces a variety of structural cardiac changes secondary to hemodynamic alterations. The increased metabolic demand imposed by the expanded adipose tissue and increased fat-free mass causes hyperdynamic circulation with increased blood volume (13, 14). Alexander et al. (13) demonstrated a positive linear correlation between the amount of overweight and both blood volume and cardiac output. Adipose tissue blood flow averages 2 to 3 mL min^{-1} 100 g^{-1} under resting conditions (15). Thus, 100 kg of fat would require as much as 3 L/min blood flow to fulfill the requirement of adipose tissue flow (14,15). These changes may increase intracardiac pressures (16). BMI emerged as a significant independent predictor of heart failure in both sexes. Approximately 11% of cases of heart failure among men and 14% among women in the community could be attributable to obesity alone (17).

However, when we analyzed BNP, we did not detect differences in plasma levels compared to the control group. Wang et al. (17) observed a progressive decrease in plasma natriuretic peptides with increasing BMI. Similar to our results, Hanusch-Enserer et al. (18) found higher levels of NT-proBNP in morbidly obese subjects, with a significant decrease during weight loss after laparoscopic-adjustable gastric banding. Hermann-Arnhof et al. (19), analyzing NT-proBNP as an indicator of possible cardiovascular disease in severely obese individuals and comparing it with that of patients in different stages of heart failure, concluded that NT-proBNP was increased in obese individuals and the levels were similar in patients with NYHA class I heart failure. Our results were similar to those

obtained by Wang et al. (17) regarding BNP levels, which were not increased in level III obese patients.

Previous studies on BNP and NT-proBNP within a large, heterogeneous population of patients with suspected cardiovascular disease at risk of cardiovascular dysfunction have suggested that NT-proBNP might be a more discerning marker of early cardiac dysfunction than BNP (20,21). Therefore, it is useful to use NT-proBNP instead of BNP to evaluate cardiac dysfunction in obese individuals.

NT-proBNP and BNP are secreted by the myocardium on an equimolar basis in cases of volume overload and increased wall tension. The elevated circulating NT-proBNP levels do not support the fact of diminished myocardial release or impaired synthesis for low BNP levels, suggesting that adipose tissues are intimately related to the natriuretic clearance receptor (17).

The diagnosis of heart failure in obese patients is difficult because such patients often have very limited mobility and may appear to be asymptomatic, even when they have significant cardiovascular disease. Moreover, in some patients the symptoms of heart failure are misdiagnosed as symptoms of obesity. The signs of left and right failure may be difficult to elicit in morbidly obese subjects and complementary examinations like echocardiography may also be difficult to perform due to a poor acoustic window (21).

The use of a noninvasive complementary method like NT-proBNP to diagnose myocardial overload in these patients may provide additional information regarding the risk of heart failure. Noninvasive imaging examinations may only identify cardiac damage later. We believe that an earlier diagnosis of cardiac burden due to obesity could avoid the subsequent evolution to myocardial dilation and to obesity cardiomyopathy.

Duration of obesity is also an important determinant of LV systolic function. It correlates positively with LV end-systolic stress, LV dimension in diastole, and LV mass/height index. It also contributes to impairment in left diastolic function. We observed higher NT-proBNP levels in patients with a longer duration of obesity. The clinical syndrome of obesity cardiomyopathy has been described in patients with a duration of obesity longer than 10 years (6,7).

Table 2. Spearman correlation between Log-NT-proBNP and obesity variables (N = 33).

Variable	NT-proBNP	
	r	P value
Waist circumference	-0.275	0.14
Duration of obesity	0.339	<0.004
Body mass index	0.006	0.97

NT-proBNP = N-terminal-pro-brain natriuretic peptide.

We did not observe a correlation between NT-proBNP and the presence of hypertension. However, Wang et al. (17) noticed lower BNP levels in hypertensive obese patients compared to non-hypertensive individuals. Similar to our results, Grandi et al. (22) observed that plasma levels of BNP were not significantly different between normotensive and hypertensive obese subjects with the same morpho-functional characteristics.

NT-proBNP, but not BNP, may be useful as an early diagnostic tool for the detection of cardiac burden due to severe obesity.

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