N-Terminal Pro-B-Type Natriuretic Peptide after High-Dose Chemotherapy: A Marker Predictive of Cardiac Dysfunction?

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Background: Chronic cardiac dysfunction may develop after administration of aggressive chemotherapy, sometimes leading to development of congestive heart failure (CHF). Recently, N-terminal pro-B-type natriuretic peptide (NT-proBNP) was implicated as a marker of CHF. In this study we evaluated the predictive role of NT-proBNP in patients treated with high-dose chemotherapy (HDC).

Methods: NT-proBNP was measured after 62 chemotherapy treatments in 52 patients affected by aggressive malignancies. Blood samples were drawn before the start of HDC, at the end of HDC administration, and 12, 24, 36, and 72 h thereafter. In these patients, echocardiograms were performed regularly during a 1-year follow-up.

Results: Seventeen patients (33%) had persistently increased NT-proBNP, 19 patients (36%) had only transient increases (concentrations went back to baseline at 72 h), and 16 (31%) had no increases [mean (SD) values at 72 h, 1163 (936) ng/L vs 185 (101) ng/L vs 39 (19) ng/L, respectively; \( P < 0.0001 \)]. Only patients with persistently increased NT-proBNP had a significant worsening of the left ventricular diastolic indexes from baseline to 12 months [ratio of peak early to peak late flow velocities from 1.42 (0.33) to 0.78 (0.11); \( P < 0.0001 \); isovolumetric relaxation time from 90 (15) to 141 (26) ms; \( P < 0.0001 \); E-wave deceleration time from 162 (17) to 224 (32) ms; \( P = 0.0004 \) and of the left ventricular ejection fraction [from 62.8 (3.4)\% to 45.6 (11.5)\%; \( P < 0.0001 \)].

Conclusions: Persistently increased NT-proBNP early after administration of HDC is strongly associated with development of cardiac dysfunction. This finding has important implications for identifying patients at risk of developing chemotherapy-induced cardiotoxicity.

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The risk of developing cardiac dysfunction after administration of high-dose chemotherapy (HDC)5 for the treatment of aggressive malignancies, both solid and hematologic, is well known. Use of HDC is limited by a cumulative dose-dependent cardiotoxicity, which can cause cardiomyopathy, possibly leading to congestive heart failure (CHF), particularly when chemicals of the anthracycline group are used (1, 2).

Whereas early and acute cardiotoxicities occur during or soon after treatment (3), the chronic form, which is the most serious form of cardiotoxicity, may appear months or even years after the end of chemotherapy, usually leading to the development of CHF and possibly death (4–6). As a consequence, guidelines have been published suggesting that patients at high risk of cardiotoxicity should be monitored regularly with serial echocardiography (ECHO) or with radionuclide ventriculography (7–10). However, these techniques are only partially reliable and available and are quite expensive. Moreover, they do not have the desired sensitivity for detection of early

*Nonstandard abbreviations: HDC, high-dose chemotherapy; CHF, congestive heart failure; ECHO, echocardiography; ANP, atrial natriuretic peptide; NT-proANP, N-terminal proANP; BNP, B-type natriuretic peptide; NT-proBNP, N-terminal proBNP; LVEF, left ventricle ejection fraction; LV, left ventricle; E:A ratio, ratio of peak early to peak late flow velocities; DT, E-wave deceleration time; and IVRT, isovolumetric relaxation time.
cardiac dysfunction, which may allow the administration of cardioprotective drugs (11).

Natriuretic peptides such as atrial natriuretic peptide (ANP), N-terminal proANP (NT-proANP), B-type natriuretic peptide (BNP), and N-terminal proBNP (NT-proBNP) are a family of structurally related peptides that have recently emerged as biomarkers potentially useful in the diagnosis and prognostic stratification of patients with CHF (12–15). In response to cardiac overload, ANP is released from the atrium, whereas BNP is released from the ventricle.

Only a few reports (16–25) have been published addressing the role of ANP, NT-proANP, and BNP in patients treated with chemotherapy, and no reports have been published regarding the potential role of NT-proBNP in patients treated with HDC and peripheral blood stem cell support (26). We therefore evaluated the possible predictive role of NT-proBNP in 52 patients treated with HDC, in whom ECHO was performed regularly during a 1-year follow-up.

Materials and Methods

Study Population

The study was conducted retrospectively on 52 patients treated with HDC for aggressive malignancy from January 2000 to December 2000 and from July 2002 to August 2003 in our Institute. The stocking of serum aliquots had to be stopped for a period because of a shortage of freezer space. Stocked biological specimens serially collected before and during HDC administration from 106 patients treated at our institute from January 2000 to August 2003 were potentially available for NT-proBNP determinations. Of these patients, however, only 52 (49%) had cardiac evaluations done at treatment initiation and 4 and 12 months after treatment. The specimens of all 52 patients were subsequently assayed for NT-proBNP to assess possible associations with cardiac monitoring findings.

The study population therefore consisted of a cross-sectional group of patients who received HDC for treatment of their malignancies before the onset of abnormal cardiac findings. The NT-proBNP response pattern during HDC treatment was assessed retrospectively by use of stocked specimens; cardiac indexes were then compared across NT-proBNP response pattern groups. A person blinded to the cardiac data performed the NT-proBNP assays.

The final study population consisted in 17 men and 35 women [mean (SD) age, 47 (11) years], who had a total of 62 different episodes of chemotherapy. HDC exclusion criteria were a history of ischemic, valvular, or hypertensive heart disease; uncontrolled hypertension; and left ventricular ejection fraction (LVEF) <50%. All patients except 2 had normal renal (creatinine serum concentration <80 μmol/L in women and <107 μmol/L in men) and liver function (assessed by aspartate aminotransferase and bilirubin concentrations).

The patients were treated with different drug combinations according to various schemes based on our Institute’s medical oncology protocols. No difference among drugs and NT-proBNP behavior was noticed throughout the follow-up. All drugs were administered by continuous intravenous infusion via a central venous catheter. Before HDC, progenitor cells were collected from peripheral blood after mobilization by growth factors. All patients underwent re-infusion of autologous peripheral blood progenitor cells during each course of HDC (26). Informed consent to use blood samples for research purposes was obtained at hospital admission.

Study Protocol

Each patient was treated with HDC every 28 days for 1–3 cycles depending on their specific chemotherapy schedules. Blood samples were taken at baseline (before each treatment), at the end of the infusion, and 12, 24, 36, and 72 h after the end of each chemotherapy cycle.

ECHO Analyses

Cardiac function was assessed by ECHO at baseline and at 4 and 12 months after the end of treatment. To evaluate left ventricular (LV) diastolic function, the following indexes were recorded: (a) peak flow velocity of early filling (E); (b) peak flow velocity of late (atrial) filling (A); (c), ratio of peak early to peak late flow velocities (E:A ratio); (d), E-wave deceleration time (DT); and (e), isovolumetric relaxation time (IVRT). LVEF was recorded as systolic function index.

Laboratory Methods

Blood samples were collected into a Monovette containing a sodium citrate solution (0.106 mol/L) with a dilution ratio after blood collection of 1 in 10 (1 part citrate and 9 parts blood). The samples were then centrifuged, and plasma was stored in aliquots at −30 °C until analysis was performed.

NT-proBNP was assayed by an electrochemiluminescence immunoassay (Elecsys NT-proBNP Test; courtesy of Roche Diagnostics, Mannheim, Germany) on a semiautomated analyzer (Elecsys-1010; Roche Diagnostics).

As cutoffs we used the values suggested by the manufacturer: 153 and 88 ng/L, respectively, for women and men ≥50 years of age; and 334 and 227 ng/L, respectively, for women and men >50 years of age.

Statistical Analysis

Median NT-proBNP was plotted against time of sample collection to describe the response patterns that were later evaluated with regard to different risks of cardiac damage. Patient characteristics by type of NT-proBNP response pattern were compared by the Fisher exact or χ² test for categorical variables and the Kruskal–Wallis test for continuous variables. Linear-mixed regression models were then used to assess whether cardiac function was dependent on type of NT-proBNP response pattern dur-
and after HDC initiation. Each of the 6 cardiac indexes studied was regressed on NT-proBNP response pattern group, time of assessment (baseline and 4 and 12 months after the end of HDC), and a group-by-time interaction term. A fourth covariate was added to each model to adjust for the cycle of chemotherapy to which the cardiac data corresponded, given than 10 of 52 patients contributed to the data set with observations after 2 consecutive cycles of chemotherapy. The estimates generated by the models took into account the correlation between repeated observations per patient. A compound symmetry correlation structure was specified for the data. Residuals were plotted (Q-Q plots) to verify the normality of the cardiac data. For graphical representation, unadjusted means and standard deviations are presented, whereas cited \(P\) values were obtained from the regression models. Unadjusted means were preferred over least-squares means in the absence of unbalanced data throughout follow-up. All analyses were conducted with SAS (SAS Institute). A 2-sided \(P\) value <0.05 was considered statistically significant.

### Results

According to the changes in NT-proBNP concentrations detected after the chemotherapy cycle considered, patients were divided into 3 groups: in the first group, group A (17 patients for a total of 21 cycles), plasma NT-proBNP increased significantly immediately after the end of the infusion, with high concentrations still present after 72 h [160 (95) ng/L at baseline; 1163 (936) ng/L after 72 h; \(P <0.00038\)]. In the second group, group B (19 patients for a total of 22 cycles), NT-proBNP increased after 12–36 h from the end of treatment but decreased toward baseline after 72 h [120 (107) ng/L at baseline; 185 (101) ng/L after 72 h; \(P =0.002\)]. In the third group, group C (16 patients for a total of 19 cycles), NT-proBNP decreased from baseline to 72 h [90 (95) ng/L at baseline; 39 (19) ng/L after 72 h; \(P =0.04\); Fig. 1]. Patient characteristics by NT-proBNP response pattern are summarized in Table 1.

There were no significant differences among the 3 groups regarding age, sex, and underlying malignancies, although the median age was somewhat higher in group A (54 years) than in groups B and C (47 and 44 years, respectively; \(P =0.11\)).

A total of 276 plasma NT-proBNP measurements were

### Table 1. Clinical characteristics of the study population according to NT-proBNP group.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Total</th>
<th>Group A (n = 17)</th>
<th>Group B (n = 19)</th>
<th>Group C (n = 16)</th>
<th>(P)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Median (range) age, years</td>
<td>54 (25–63)</td>
<td>35/17</td>
<td>14/3</td>
<td>12/7</td>
<td>0.11&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>Sex, F/M</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.25&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Diagnosis, n</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>0.14&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Breast cancer</td>
<td>14</td>
<td>6</td>
<td>8</td>
<td>7</td>
<td></td>
</tr>
<tr>
<td>Lymphoma</td>
<td>5</td>
<td></td>
<td>6</td>
<td>8</td>
<td></td>
</tr>
<tr>
<td>Other</td>
<td>2</td>
<td></td>
<td>8</td>
<td>4</td>
<td></td>
</tr>
<tr>
<td>Myeloma</td>
<td>7</td>
<td>2</td>
<td>4</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sarcoma</td>
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<td></td>
<td></td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Uterine cancer</td>
<td>2</td>
<td></td>
<td></td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>SCLC&lt;sup&gt;d&lt;/sup&gt;</td>
<td>1</td>
<td></td>
<td></td>
<td></td>
<td>1.00</td>
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<tr>
<td>AML</td>
<td>2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Patients with data on second cycle included, n</td>
<td>4</td>
<td>3</td>
<td>3</td>
<td></td>
<td>0.91&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td>Median (range) basal creatinine, (\mu)mol/L</td>
<td>61 (35–108)</td>
<td>55.7 (29–167)</td>
<td>51 (35–85)</td>
<td></td>
<td>0.52&lt;sup&gt;f&lt;/sup&gt;</td>
</tr>
<tr>
<td>Median (range) basal LVEF, %</td>
<td>63 (57–69)</td>
<td>61 (57–68)</td>
<td>63 (57–68)</td>
<td></td>
<td>0.53&lt;sup&gt;f&lt;/sup&gt;</td>
</tr>
<tr>
<td>Median (range) LV mass, g&lt;sup&gt;7&lt;/sup&gt;</td>
<td>151 (80–187)</td>
<td>137 (91–206)</td>
<td>147 (108–200)</td>
<td></td>
<td>0.93&lt;sup&gt;g&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup> Group A, patients with persistent NT-proBNP increase; group B, patients with only transient NT-proBNP increase; group C, patients with no NT-proBNP increase.

<sup>b</sup> Kruskal–Wallis test.

<sup>c</sup> Fisher exact test.

<sup>d</sup> SCLC, small cell lung cancer; AML, acute myeloid leukemia.

<sup>e</sup> \(\chi^2\) test.

<sup>f</sup> LV mass was calculated by use of a cubic equation at end diastole [35].

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**Fig. 1. NT-proBNP concentrations at the different sampling times.** The curves show 3 different patterns according to the changes in NT-proBNP concentration [medians and interquartile ranges (error bars)]. ●, group A; ■, group B; ●, group C.
performed during 62 treatment cycles in 52 patients. For 10 patients, the blood samples after 2 chemotherapy cycles were available. Among these, 6 had NT-proBNP measurements done during cycles 1 and 2, 2 had measurements done during cycles 1 and 3, and 2 had measurements done during cycles 2 and 3. We detected no differences between 2 curves for the same patient. The data set analyzed included 26, 21, and 15 curves corresponding to cycles 1, 2, and 3, respectively. Nine of 52 patients (17.3%) had NT-proBNP values at baseline greater than the cutoff, 3 in each group.

LV diastolic values were available for 42 patients (14 patients in group A, 15 in group B, 13 in group C). The mean E:A ratio decreased significantly over time in group A, whereas it remained unchanged in the other 2 groups [in group A, 1.42 (0.33) at baseline, 0.97 (0.25) at 4 months, and 0.78 (0.11) at 12 months; in group B, 1.29 (0.34) at baseline, 1.17 (0.33) at 4 months, and 1.17 (0.31) at 12 months; in group C, 1.25 (0.26) at baseline, 1.22 (0.28) at 4 months, and 1.24 (0.25) at 12 months; significant group-by-time interaction, \( P < 0.0001 \); Fig. 2]. DT and IVRT increased significantly during follow-up in group A, whereas the changes were not significant in groups B and C (Table 2). LV systolic values were available for all patients. LVEF was significantly decreased only in the group A patients, with the mean value decreased below the threshold of 50% after 12 months [62.8 (3.4)% at baseline, 54.4 (6.6)% at 4 months, and 45.6 (11.5)% at 12 months; \( P < 0.0001 \); proportion of patients with value <50% = 10 of 17, or 59% (95% confidence interval, 36%–82%)]. Among these, 4 patients reported an important decrease in dyspnea threshold, without overt signs of heart failure. None of the patients in the other 2 groups had LVEF values <55% (Fig. 2).

**Discussion**

Our results suggest that persistently increased plasma NT-proBNP after HDC administration is associated with the development of subsequent cardiac dysfunction. Cardiac toxicity is a well-known complication of several antineoplastic treatments, in particular when anthracyclines are used as part of the chemotherapy regimen (27).

In addition to anthracyclines, alkylating agents such as cyclophosphamide, cisplatin, busulfan, and mitomycin have also been associated with cardiotoxicity, as have paclitaxel, etoposide, vinca alkaloids, fluorouracil, and other drugs (28). The most serious clinical problem is represented by progressive heart failure and other cardiac

| Table 2. Mean (SD) DT and IVRT in patients grouped according to NT-proBNP response pattern during HDC. |
|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
| **Group** | **DT, ms** | **IVRT, ms** | **DT, ms** | **IVRT, ms** | **DT, ms** | **IVRT, ms** |
| **Baseline** | **4 months** | **12 months** | **P** | **Baseline** | **4 months** | **12 months** | **P** |
| C (n = 13) | 178 (40) | 189 (45) | 188 (38) | NS* | 97 (14) | 101 (14) | 94 (18) | NS |
| B (n = 15) | 171 (35) | 188 (63) | 198 (42) | NS | 94 (14) | 97 (11) | 91 (16) | NS |
| A (n = 14) | 162 (17) | 201 (28) | 224 (32) | <0.0001 | 90 (15) | 123 (24) | 141 (26) | <0.0001 |

*NS, not significant.
events developing from weeks to years after anthracycline therapy (29, 30).

Although currently available noninvasive diagnostic techniques such as ECHO and radionuclide imaging may reveal cardiac dysfunction before it becomes clinically overt, they lack adequate sensitivity to detect early cardiac damage and to identify individuals at increased risk of subsequent CHF while they are still receiving chemotherapy (7–10). Hence, there is the need to look for easy, sensitive, moderate-cost methods, such as serum tests, to predict chemotherapy-induced cardiotoxicity.

Natriuretic peptides have been demonstrated to be useful markers of LV dysfunction in both symptomatic and asymptomatic patients. High concentrations of plasma natriuretic peptides have been described not only in patients with acute myocardial infarction or advanced CHF but also in patients with asymptomatic or minimally symptomatic LV dysfunction (31, 32). Only a few published reports have evaluated the role of natriuretic peptides in chemotherapy-treated patients. Some studies concluded that there is an association between LV dysfunction and increased natriuretic peptide concentrations (16–21). Only a few anecdotal reports (22–25) exist on the potential role of ANP and/or BNP as markers useful for detecting subclinical cardiac dysfunction, whereas no reports have been published on the role of NT-proBNP.

The present study evaluates for the first time the usefulness of NT-proBNP as an early marker predictive of cardiac dysfunction in patients affected by aggressive malignancies treated with HDC. We found 3 distinct NT-proBNP concentration patterns. Some patients had no changes in NT-proBNP concentrations over the 6 sample times, whereas another group had only a transient increase, with concentrations normalizing at 72 h. In these patients, no significant ECHO index alterations were recorded during follow-up. Only patients with persistently increased NT-proBNP concentrations that remained increased at 72 h after the end of HDC administration developed some form of cardiac impairment during the 12 months of observation, with a significant association between worsening of both diastolic and systolic values and persistently increased NT-proBNP. In particular, the ECHO monitoring revealed increases in DT and IVRT, an E:A ratio change from >1 to <1 over time, and a decrease in LVEF that were more pronounced after 12 months. The decrease in the mean LVEF from 62.8% to 45.6%, with 10 of the patients (59%) having values <50%, represents the clearest indicator of cardiac function impairment. Moreover, 5 of 7 of the remaining patients in this group had a >10% decrease in LVEF (13%–21%); even if LVEF remained within the limits of normality, it is likely that this decrease may represent a risk factor.

Our findings confirm previous reports of a close relationship between natriuretic peptides and cardiac dysfunction in patients treated with anthracycline (16–21). However, this is the first report suggesting that early increases in NT-proBNP concentrations, measured just after the end of a chemotherapy treatment and well before any clinical or ECHO manifestation, may be considered as an early marker of cardiac involvement. The persistence of increased concentrations for more than 72 h suggests that in these patients the increase in NT-proBNP reflects the presence of an underlying reduced functional myocardial reserve or reduced cardiac tolerance to cardotoxic agents. In fact, the myocardial damage induced by HDC might elicit a neurohormonal activation, as evidenced by persistently increased NT-proBNP.

Our study has some limitations. The first limitation is that these preliminary results were obtained from a retrospective analysis including a reduced number of patients. However, the evaluation of plasma NT-proBNP concentrations was done without knowledge of the results of the ECHO performed during the follow-up period of 1 year, and there were no patients in whom signs of cardiac dysfunction occurred without a previous persistent increase in NT-proBNP concentrations. Nine patients had NT-proBNP concentrations slightly above the reference value at baseline; these patients were equally distributed among the 3 groups, and we focused particularly on the changes in NT-proBNP concentration over the different sampling times rather than on the absolute NT-proBNP values. Lastly, we did not evaluate the concentrations of other natriuretic peptides because of a lack of the necessary equipment in our laboratory. We have already demonstrated that troponin I is an early and useful marker of chemotherapy-induced cardiotoxicity (30, 33, 34). Further prospective studies are needed to clarify whether both markers give the same kind of information or whether their combination may better help in identifying persons at high risk.

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References