Increased levels of various neurohumoral factors have been found in patients with congestive heart failure (CHF), and high plasma concentrations of norepinephrine (NE), renin, endothelin-1, atrial natriuretic peptide, and brain natriuretic peptide (BNP) have been reported as significant prognostic predictors, suggesting that neurohumoral activation is important in the pathogenesis of CHF. In particular, activation of the sympathetic nervous system has an important role in the pathophysiology of CHF; that is, cardiac stores of NE are depleted, neuronal NE release is increased and adrenoreceptors are desensitized in CHF.

Cardiac imaging with 123I-metaiodobenzylguanidine (MIBG), an analog of NE, is now used to noninvasively assess sympathetic nerve function and has improved in patients with CHF after treatment with angiotensin-converting enzyme (ACE) inhibitors and ß-blockers suggesting that the impairment of cardiac adrenergic function is reversible. However, there are not any reports of whether serial MIBG imaging before and after additional treatments for DCM is useful for predicting mortality.

The plasma concentration of BNP, which is mainly derived from the ventricle, is a sensitive and major prognostic index. We reported that a high plasma concentration of BNP after 3 months of optimized treatment is an independent risk factor for mortality in patients with CHF, including those with DCM, despite improvements in left ventricular ejection fraction (LVEF) and symptoms. To our knowledge, there are not any reports on whether the plasma BNP concentration remains an independent prognostic predictor if combined with cardiac MIBG imaging.

Therefore, we investigated whether repeated measurement of the cardiac MIBG parameters before and after additional treatments can provide prognostic information independent of the clinical and neurohumoral factors (such as plasma NE and BNP concentrations) in patients with DCM.

**Methods**

**Patients**

We studied 85 consecutive patients hospitalized with DCM (59 male, 26 female; mean age, 55.4±1.4 years). All patients had a LVEF (measured by the biplane disc sum-
Table 1 Characteristics of 74 Patients With Dilated Cardiomyopathy: Survivors and Nonsurvivors or Patients With and Without a Cardiac Event

<table>
<thead>
<tr>
<th></th>
<th>All patients (n=74)</th>
<th>Survivors (n=62)</th>
<th>Nonsurvivors (n=12)</th>
<th>Cardiac event (–) (n=51)</th>
<th>Cardiac event (+) (n=23)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Age (years)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>M/F</td>
<td>55.4±1.4</td>
<td>55.8±1.6</td>
<td>53.4±2.6</td>
<td>55.1±1.9</td>
<td>56.2±1.8</td>
</tr>
<tr>
<td><strong>LVEF (%)</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Before</td>
<td>31.2±3.1</td>
<td>33.9±3.1</td>
<td>22.1±1.9</td>
<td>31.9±1.9</td>
<td>29.7±1.7</td>
</tr>
<tr>
<td>After 6 months</td>
<td>34.9±1.3</td>
<td>36.5±1.5</td>
<td>26.8±2.2**</td>
<td>37.2±1.7</td>
<td>29.4±1.8***</td>
</tr>
<tr>
<td><strong>NYHA class II/III/IV</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Before</td>
<td>7/57/10</td>
<td>7/51/4</td>
<td>0/6/6**</td>
<td>7/40/4</td>
<td>0/17/6*</td>
</tr>
<tr>
<td>After 6 months</td>
<td>63/110</td>
<td>55/70</td>
<td>8/4/0</td>
<td>46/50</td>
<td>17/00</td>
</tr>
</tbody>
</table>

LVEF, left ventricular ejection fraction; NYHA, New York Heart Association; Cardiac event, cardiac death (worsening CHF or sudden death), hospitalization for worsening CHF and lethal arrhythmia; CHF, congestive heart failure. *p<0.05 vs cardiac event (–); **p<0.01 vs survivors; ***p<0.01 vs cardiac event (–).

Table 2 Optimal Treatments for Dilated Cardiomyopathy in 74 Patients With CHF During 6 Months

<table>
<thead>
<tr>
<th></th>
<th>All patients before / after 6 months</th>
<th>Survivors (n=62)</th>
<th>Nonsurvivors (n=12)</th>
<th>Cardiac event (–) (n=51)</th>
<th>Cardiac event (+) (n=23)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Diuretics</strong></td>
<td>63 (85%) / 66 (89%)</td>
<td>51/54</td>
<td>12/12</td>
<td>41/43</td>
<td>22/23</td>
</tr>
<tr>
<td><strong>Digitalis</strong></td>
<td>49 (66%) / 51 (69%)</td>
<td>39/40</td>
<td>10/11</td>
<td>32/33</td>
<td>17/18</td>
</tr>
<tr>
<td><strong>ACE inhibitors</strong></td>
<td>20 (27%) / 59 (80%)</td>
<td>13/48</td>
<td>7/11</td>
<td>11/38</td>
<td>9/21</td>
</tr>
<tr>
<td><strong>β-blocker</strong></td>
<td>15 (20%) / 63 (85%)</td>
<td>12/54</td>
<td>3/9</td>
<td>9/45</td>
<td>6/18</td>
</tr>
<tr>
<td><strong>Vasodilator</strong></td>
<td>8 (11%) / 3 (4%)</td>
<td>7/2</td>
<td>1/1</td>
<td>6/2</td>
<td>2/1</td>
</tr>
</tbody>
</table>

ACE, angiotensin-converting enzyme; Cardiac event, cardiac death (worsening CHF or sudden death), hospitalization for worsening CHF and lethal arrhythmia; CHF, congestive heart failure.

Table 2 Optimal Treatments for Dilated Cardiomyopathy in 74 Patients With CHF During 6 Months

MIBG Imaging
All patients received 111 MBq (3 mCi) of $^{123}$I-MIBG intravenously and the images were obtained by a single-head gamma camera with a low-energy collimator (Toshiba GCA-901A). Energy discrimination was provided by a 20% window centered on the 159 keV photopeak. A 5-min static acquisition was performed at 15 min (early) and 180 min (delayed) after radioisotope injection as previously reported. On the anterior planar image, a region of interest (ROI) was manually placed over the heart and another ROI was placed over the upper mediastinal area. From the early and delayed images, the heart to mediastinum (H/M) ratio and the washout rate were calculated from the average counts in each ROI by 2 independent observers. The washout rate was defined as the percentage change in cardiac activity (H) from the early to the delayed image within the left ventricular area:

$$\text{Washout rate} = \frac{\text{[(H) – (M)] early – [(H) – (M)] delayed}}{[(H) – (M)] early} \times 100 \%.$$  

Decay correction was not done. The interobserver differences for the H/M ratio (delayed) and washout rate were not significant. The normal ranges of the H/M ratio (delayed) and washout rate were 2.60±0.24 and 28.0±3.1%, respectively.

Measurement of Plasma Neurohumoral Factors
Samples for the assay of plasma BNP concentration were...
transferred to chilled disposable tubes containing aprotinin (500 kallikrein inactivator units/ml) and immediately placed on ice and centrifuged at 4°C. The plasma BNP concentration was measured with a specific immunoradiometric assay using a commercial kit (Shionogi, Osaka, Japan) as reported previously. Briefly, this assay uses 2 monoclonal antibodies against human BNP, one recognizing a carboxy-terminal sequence and the other the ring structure of BNP, and measures BNP by sandwiching it between the 2 antibodies without the need for plasma extraction. The plasma NE concentration was measured by high-performance liquid chromatography as reported previously.

Statistical Analysis
All data are reported as the mean±SEM. Using Student's paired t-test, the H/M ratio (delayed), washout rate, BNP and NE were compared before and after optimized treatment. Using repeated measures, serial changes in the MIBG parameters and neurohumoral factors were compared between survivors and nonsurvivors. Categorical data were compared using the chi-square test. The prognostic values of 19 variables were tested by Cox proportional hazards regression analysis. The Kaplan-Meier method was used to calculate the cumulative survival in groups of patients that were divided according to the median value of the H/M ratio (delayed) and BNP, and in groups of patients that showed a change in the H/M ratio (delayed). Differences between survival curves were assessed by the log-rank test. The diagnostic accuracy of the H/M ratio (delayed) before treatment, the changes in the H/M ratio (delayed) and the plasma BNP concentration after 6 months for prediction of mortality was also investigated by using receiver operating characteristic (ROC) curves. The areas under the curves were calculated and analyzed using a one-tailed test. Statistical significance was defined at p<0.05.

Results
Outcome and Patient Characteristics
Eighty-five patients were enrolled; 4 died of worsening CHF, 5 were hospitalized for worsening CHF within the 6 months of optimized treatment, and 2 patients dropped out, so 74 patients could be followed-up after additional treatment (Table 1). A total of 23 cardiac events were observed during a mean follow-up of 731±65 days: 12 cardiac deaths and 11 patients who were hospitalized for worsening of CHF or for severe arrhythmia. There was a significant difference in NYHA functional class between the survivors and nonsurvivors at baseline, but none in age, gender or LVEF. Similarly, only the NYHA functional class differed between the patients who had a cardiac event and those who did not. After additional treatment, there was a significant difference in the LVEF between the survivors and nonsurvivors.

Fig 1. Comparison of [123I]-metaiodobenzylguanidine (MIBG) parameters and neurohumoral factors before and after 6 months of optimized additional treatments in patients with dilated cardiomyopathy. *p<0.05 vs before treatment, **p<0.01 vs before treatment. H/M, heart to mediastinum; BNP, brain natriuretic peptide; NE, norepinephrine.

Fig 2. (Left) Changes in the heart to mediastinum (H/M) ratio and washout rate between before and 6 months after optimized additional treatment in nonsurvivors (●). (Right) Changes in the H/M ratio and washout rate between before and 6 months after optimized additional treatment in survivors (●). ●, mean value in nonsurvivors. ●, mean value in survivors. *p<0.01 vs before additional treatment for dilated cardiomyopathy.

Fig 3. Delayed MIBG images obtained in a survivor (Top) and a nonsurvivor (Bottom). On the left of each panel are images before optimized treatment, and on the right are images 6 months later. The H/M ratio in the survivor increased from 1.92 to 2.46, whereas that in the nonsurvivor decreased from 1.97 to 1.72.
Cardiac MIBG Parameters and Neurohumoral Factors Before and After Additional Treatments

The mean H/M ratio (delayed) in all subjects was significantly increased after 6 months of additional treatments (1.89±0.03 before vs 2.00±0.04 after) (Fig 1). There was no difference in the baseline H/M ratio (delayed) between survivors and nonsurvivors. In survivors, the H/M ratio (delayed) was significantly increased after 6 months of additional treatment compared with the nonsurvivors in whom it was significantly decreased (11 of 12 nonsurvivors; Fig 2). Fig 3 shows representative MIBG images of survivors and nonsurvivors. The mean washout rate in all subjects was significantly increased after 6 months of additional treatment compared with the nonsurvivors in whom there was a slight increase in nonsurvivors (Fig 2).

The plasma concentrations of BNP and NE in all subjects after 6 months of additional treatment were significantly improved compared with those at baseline (BNP, 275±36 vs 202±29 pg/ml; NE, 422±32 vs 362±25 pg/ml), as shown in Fig 1. However, in nonsurvivors, the plasma concentrations of BNP and NE were high both before and after 6 months of additional treatment (BNP, 637±113 vs 574±99 pg/ml; NE, 677±150 vs 585±225 pg/ml).

Univariate and Multivariate Predictors of Mortality and Morbidity

Nineteen variables were analyzed using univariate and stepwise multivariate Cox proportional hazards regression analysis for mortality and morbidity (Table 3). By univariate analysis, NYHA (before treatment and at 6 months), LVEF (at 6 months), absolute change in the LVEF, BNP (before and at 6 months), NE (before and at 6 months), H/M ratio (delayed) (at 6 months), absolute change in the H/M ratio (delayed), washout rate (at 6 months) and absolute change in the washout rate were significant predictors of mortality. By stepwise multivariate analysis, only the degree of the absolute change in the H/M ratio (delayed) at 6 months compared with that before optimized treatment, and a high concentration of plasma BNP (at 6 months) were significant independent predictors.

Furthermore, the same factors were analyzed using univariate and stepwise multivariate Cox proportional hazards regression analysis for mortality and morbidity. NYHA (at 6 months), LVEF (at 6 months), absolute change in the LVEF, BNP (before treatment and at 6 months), NE (before treatment and at 6 months) and MIBG parameters were significant predictors of mortality and morbidity. By stepwise multivariate analysis, the degree of the absolute change in the H/M ratio (delayed) at 6 months compared with that before optimized treatment, and a high concentration of plasma BNP (at 6 months) were independent predictors.

Kaplan-Meier Lifetime Analysis

The patients were stratified into 2 groups according to the change in the H/M ratio (delayed), and also according to the median value of the H/M ratio (1.89) before and after (1.98) 6 months of optimized treatment and the plasma BNP (92 pg/ml) after 6 months of optimized treatment. Cumulative survival curves were constructed according to the Kaplan-Meier method (Fig 4). The group with a decrease in the H/M ratio showed a significantly lower survival rate, as did the group with a median H/M ratio <1.89 after treatment and <1.98 after optimized treatment. Moreover, the group with a median plasma BNP level ≥92 pg/ml had a significantly lower survival rate.

Accuracy of MIBG Parameters and BNP as Predictors of Mortality

The ROC curves showed a greater area under the curve for the plasma BNP concentration at 6 months and for the absolute change in the H/M ratio than for the H/M ratio before treatment. ROC analysis also showed no significant differences between the absolute change in the H/M ratio and the plasma BNP concentration after 6 months of opti-
The sensitivity and specificity of mortality of the patients with a decrease in the H/M ratio after compared with before optimized treatment were 92% and 73%, respectively. The sensitivity and specificity of mortality of the patients with a plasma concentration of BNP (after 6 months of optimized treatment) >170 pg/ml were, respectively, 83% and 81% (the optimal cut-off point for predicting mortality of the patients with a decrease in the H/M ratio and brain natriuretic peptide (BNP) (92 pg/ml) after 6 months of optimized additional treatment in patients with DCM.

Usefulness of the Change in the MIBG Parameters After Optimized Treatments

Altered adrenergic function is a hallmark of CHF and cardiac 123I-MIBG imaging is a useful tool for evaluating the prognosis and response to treatment in patients with CHF.20–26 Although the parameters of cardiac 123I-MIBG imaging improve after treatment for CHF,24–26 it was not known whether serial measurements of MIBG imaging before and after additional treatments for DCM were useful for predicting mortality and we have shown from the results of the present study that an absolute change in the H/M ratio is an independent prognostic predictor in patients with DCM, thus reinforcing the importance of repeat MIBG imaging (measuring the H/M ratio) in patients with CHF caused by DCM.

There was no significant difference in the H/M ratio between survivors and nonsurvivors in the present study before additional treatment, although others have reported that the H/M ratio differs significantly between survivors and nonsurvivors.21,32 This discrepancy may be caused by differences in the treatment period or the small number of patients in the present study. Cohen-Solal et al reported that there was no significant difference in H/M ratio between survivors and nonsurvivors or cardiac transplantation recipients, which concurs with our results, so multicenter research involving larger numbers of patients is needed to clarify the significance of the findings.

Our results showed that the mean value of both the H/M ratio and the washout rate were significantly improved after optimized treatments, which is consistent with previous reports.24–26 However, in nonsurvivors, the H/M ratio was significantly decreased and the washout rate did not improve after additional treatment with ACE inhibitors and/or β-blockers, which suggests that the prognosis may be poor for those patients for whom MIBG imaging does not show an improvement after additional treatment. In other words, DCM patients whose H/M ratio is increased or unchanged after 6 months of optimized treatment have a low risk of death regardless of the baseline value of the H/M ratio. Decreased MIBG uptake (H/M ratio) may reflect excessive neuronal NE release, impaired storage vesicles or impaired neuronal re-uptake function at presynaptic nerve terminals. Patients whose H/M ratio decreased after 6 months of optimized treatment may have had an increase in the NE concentration at the synaptic level that induced deleterious effects such as myocyte calcium overload, arrhythmias or myocardial injury. These mechanisms may account for the poor prognosis for patients with DCM predicted by the decrease in H/M ratio in the present study.

With regard to the MIBG parameters, both the H/M ratio and washout rate after 6 months of optimal treatment were significant prognostic factors, although a change in the H/M ratio was the strongest predictor of all. Therefore, the H/M ratio is more useful for predicting the mortality of patients with DCM than the washout rate, which is consistent with the results of previous studies although there have been contradictory reports.32,33 The washout rate is
measured from using data obtained from the early and delayed images; however, the early image includes a small component of extraneuronal uptake and, if MIBG uptake in the myocardium has already been depleted in a large area, NE release from the myocardium may not increase markedly. These considerations may explain why the washout rate was not as good a prognostic factor as the change of H/M ratio (delayed) and the clinical usefulness of these MIBG parameters remains controversial.

In contrast to the MIBG parameters, the plasma concentration of NE before or after 6 months of additional treatment was not an independent prognostic predictor.

LVEF is also an important index for evaluating the prognosis or treatment effect in patients with CHF and is currently widely used to assess the response to \( \beta \)-blocker therapy. In the present study, an absolute change in the LVEF was not found to be an independent predictor of mortality in multivariate analysis, in contrast to an absolute change in the H/M ratio. This result suggests that the MIBG imaging parameters are a more sensitive marker for predicting the prognosis in heart failure treatment than LVEF.

Comparison of MIBG Parameters and Plasma BNP as Prognostic Predictors

Both the plasma BNP concentration and MIBG uptake have been reported as important prognostic predictors, and their prognostic value has been compared, although only with one-point measurement. These parameters reportedly change in response to treatments, but until now, a comparison of serial measurement of MIBG parameters and plasma BNP concentration before and after additional treatments for the purposes of assessing prognosis has not been performed. The present study demonstrated that a worsening of the H/M ratio and a high concentration of plasma BNP after treatment were significant independent predictors of mortality in multivariate analysis.

The ROC curves showed a greater area under the curve for the plasma BNP at 6 months and for the change in the H/M ratio after treatment and ROC analysis showed no significant differences between the change in the H/M ratio (delayed) (H/M 6 months – H/M before) or plasma BNP at 6 months. The diagnostic accuracy for predicting mortality was equal for a decrease in the H/M ratio and the plasma BNP concentration after 6 months of treatment, suggesting that information from MIBG imaging together with the plasma BNP concentration is important for predicting mortality. These 2 parameters (MIBG, a radionuclide marker of myocardial sympathetic function and BNP, a biochemical marker of hemodynamic abnormality and/or myocardial injury) may be useful for identifying potential high-risk patients who have been treated with standard therapy including ACE inhibitors and \( \beta \)-blockers.

Study Limitations

We focused on patients with DCM and excluded patients with ischemic heart disease for 2 reasons. Experimental studies have demonstrated that myocardial ischemia may cause extensive myocardial catecholamine depletion and adrenergic denervation in the infarcted area. Thus, in the ischemic myocardium, the decrease in MIBG uptake may be more closely related to the extent of the ischemia than to the deterioration of contractile function. Moreover, in patients with ischemic heart disease, revascularization is obviously the parameter with the most important prognostic consequences.

All patients in the present study were treated with additional drugs such as digitalis, diuretics, ACE inhibitors or carvedilol for 6 months as prescribed independently by their physicians. Because of side effects, such as hypotension, dry cough or worsening of renal dysfunction, not all patients could be treated with ACE inhibitors, and carvedilol was not always given to patients who showed improvement of CHF symptoms after other treatments such as diuretics or ACE inhibitors. Otherwise, the optimized treatments were not randomized, so it would be difficult to evaluate the effects of specific drugs for CHF on mortality. If we evaluated drug treatments after 6 months of optimized treatment in addition to the other 19 variables on Cox multivariate analysis, none of the drugs would have been independent predictors of morbidity.

Moreover, we did not assess the H/M ratio (early) because it may not precisely reflect presynaptic uptake; previous reports could not show a significant difference in the early myocardial uptake between patients with DCM and control subjects.

Finally, although the relatively small number of deaths in this study was also a limitation, our findings showed that comparison of the repeat measurements of MIBG parameters and biochemical markers in the same patient can provide important information for evaluating the pathophysiology of CHF and the effects of treatment.

Conclusions

Evaluating the serial changes in the H/M ratio on cardiac MIBG, which reflect sympathetic nervous activity, and the plasma BNP concentration, which reflects hemodynamic abnormality and/or left ventricular damage, provides important information about the response to treatment and the prognosis of patients with CHF.

Acknowledgments

We thank Dr Shinzaro Yoshida, Dr Shinro Matsuo, and the nuclear medicine staff at Shiga University of Medical Science. We also wish to thank Ms Rako Sakaguchi for her excellent technical assistance.

References

Cardiac MIBG and BNP in CHF