Restrictive Diastolic Abnormalities Identified by Doppler Echocardiography in Patients With Thalassemia Major

Paolo Spirito, MD, Gabriele Lupi, MD, Caterina Melevendi, MD, and Carlo Vecchio, MD

The consequences of transfusional iron overload on left ventricular diastolic filling have never been investigated systematically in patients with thalassemia major. In the present study, the pattern of left ventricular filling was assessed by Doppler echocardiography in 32 patients with thalassemia major (age, 17±5 years) who had not experienced symptoms of heart failure and had normal left ventricular systolic function. Data were compared with those obtained in 32 age-matched and sex-matched normal subjects. An abnormal Doppler pattern of left ventricular filling with increased flow velocity at mitral valve opening followed by an abrupt and premature decrease of flow velocity in early diastole was identified in the patients with thalassemia. Peak flow velocity in early diastole was increased in patients compared with controls (90±10 vs. 81±15 cm/sec; p<0.01), and rate of deceleration of flow velocity after the early diastolic peak and the ratio between the early and late (atrial) peaks of flow velocity were also increased (1,050±325 vs. 762±193 cm/sec\(^2\) and 2.7±0.7 vs. 2.2±0.5, respectively; p<0.001), whereas flow velocity deceleration time was reduced (97±22 vs. 119±19 msec; p<0.001). This Doppler pattern of diastolic filling is usually described as "restrictive" and reflects a decrease in left ventricular chamber compliance. A restrictive pattern of left ventricular filling was also identified in the subgroup of 16 study patients who had undergone optimal iron chelation therapy with deferoxamine. Rate of deceleration of flow velocity after the early diastolic peak was increased, and flow velocity deceleration time was reduced in these 16 patients (1,080±356 cm/sec\(^2\) and 94±19 msec) compared with controls (795±214 cm/sec\(^2\) and 118±19 msec, respectively; p<0.01). The results of this investigation demonstrate that left ventricular filling is altered in patients with thalassemia major and that diastolic abnormalities develop in an early phase of cardiac involvement, when symptoms of heart failure are absent and systolic function is normal. The findings of this study also suggest that chelation therapy with deferoxamine does not completely protect patients with thalassemia from myocardial damage due to iron-related cardiac toxicity. (Circulation 1990;82:88-94)

In patients with thalassemia major, long-term transfusion therapy, extravasal hemolysis, and increased intestinal absorption of iron result in systemic iron overload; the deposition of iron in the myocardium causes left ventricular dysfunction.\(^1\)\(^-\)\(^2\) Although hemochromatosis of the heart is generally described as a dilated cardiomyopathy with increased left ventricular diastolic cavity dimension and depressed systolic function,\(^1\)\(^-\)\(^4\) a restrictive cardiomyopathy with impaired left ventricular filling has also been reported in some patients with idiopathic hemochromatosis.\(^5\)\(^-\)\(^7\) Diastolic filling, however, has never been systematically investigated in patients with thalassemia major.

Doppler echocardiography has been extensively used to assess left ventricular diastolic filling in a variety of cardiac diseases\(^8\)\(^-\)\(^10\) and, more recently, has been shown capable of identifying restrictive patterns of ventricular filling.\(^11\)\(^-\)\(^13\) Therefore, in the present study, we used Doppler echocardiography to assess left ventricular diastolic filling in a relatively large group of patients with thalassemia major who had not experienced symptoms of heart failure and had normal left ventricular systolic function and diastolic cavity dimension.

Methods

Selection and Characterization of Patients

Forty-three patients with β-thalassemia major (homozygous form) are currently followed in the hematology clinic of our institution. Of these 43
patients, 32 met the following criteria and were included in the present investigation: 1) absence of cardiac symptoms or clinically apparent heart disease and 2) normal left ventricular end-diastolic cavity dimension (≤54 mm) and systolic fractional shortening (≥30%) as assessed by echocardiography. Additionally, none of these 32 study patients showed left ventricular hypertrophy on the standard 12-lead electrocardiogram (according to the Romhilt-Estes criteria in patients more than 16 years of age,14 and Garson criteria15 in patients less than 16 years of age). None of the patients was treated with cardiovascular medications.

The 32 study patients were in the age range of 9–28 years (mean, 17±5 years). Each of the patients was receiving transfusions every 2–3 weeks, to maintain hemoglobin levels between 10.5 and 13.5 g/dl. In 31 of the 32 patients, transfusion therapy had been started before the age of 3 years (1.7±1 years) and, in the remaining patient, at age five. At the time of the cardiac evaluation, cumulative transfusion loads varied from 180–591 units. Each patient received a transfusion within 3 days before cardiac evaluation (mean, 1.5 days), and hemoglobin levels were determined before and after transfusion in all patients.

Of the 32 study patients, 20 started iron chelation treatment with deferoxamine before the age of 10 years (mean age, 7±5 years; range, 3–9 years), and 11 patients started chelation therapy after the age of 10 years (mean age, 16±3 years; range, 11–19 years). The remaining patient refused deferoxamine treatment. The deferoxamine infusion program was individualized in each patient, and dosages were adjusted to maintain serum ferritin below 1,300 ng/ml. Deferoxamine was delivered by subcutaneous infusion with an infusion pump for 8–12 hours (usually during the night). Deferoxamine dosages ranged at 25–50 mg/kg body wt, and were infused 4–6 days a week. The patients kept a daily diary of the amount of deferoxamine infused. Since the beginning of deferoxamine therapy, all patients were on daily oral supplementation of ascorbic acid (50–100 mg/day).

The 31 study patients who underwent iron chelation showed a variable compliance to deferoxamine therapy. Sixteen patients showed optimal compliance, defined as more than 90% adherence to the deferoxamine infusion program. Eleven other patients were considered moderately compliant, with at least 50% adherence to the infusion program. The remaining four patients underwent sporadic chelation and were considered noncompliant. The number of deferoxamine prescriptions requested by each patient during the years of treatment was also used to verify compliance with the infusion program.

Control Group

Thirty-two age-matched and sex-matched normal subjects without clinical, electrocardiographic, and echocardiographic evidence of cardiovascular disease served as controls; their age range was 9–28 years (mean, 17±5 years), and 18 (56%) were male.

Doppler Ultrasound

A 77020AI Hewlett-Packard Ultrasound System (Hewlett-Packard Co.) was used to perform the Doppler echocardiographic studies. Doppler signals were recorded on videotape simultaneously with a lead II electrocardiogram and phonocardiogram. The Doppler transmitral flow velocity profile was obtained from the apical four-chamber view, as previously described in detail.16 Doppler tracings of patients and controls were coded, and measurements were obtained by one observer who had no knowledge of the identity of the subjects.

In each subject, four to six consecutive cardiac cycles with the highest diastolic flow velocity and the steepest descent of flow velocity in early diastole, as well as the best signal-to-noise ratio, were chosen for analysis. The following measurements (Figure 1, panel A) were obtained: 1) the time from the aortic component of second heart sound to the onset of diastolic flow velocity (A2–D), a measure of duration of isovolumic relaxation17; 2) peak early diastolic flow velocity, measured as the height of the early peak of flow velocity (E); 3) rate of deceleration of flow velocity in early diastole, measured as the EF slope; 4) flow velocity deceleration time (dt), measured as the distance between the projection of the early peak E on the baseline and the point where the EF slope encounters the baseline; 5) peak flow velocity during atrial contraction, measured as the height of the late peak of flow velocity (A); and 6) ratio between the early and late peaks of flow velocity (E/A). These Doppler diastolic indexes have previously been shown to have satisfactory reproducibility.16,18 In the individual patient analysis, the diastolic filling pattern was judged to be abnormal if one or more Doppler indexes exceeded the 95% confidence limits derived from the data obtained in the 32 age-matched and sex-matched normal control subjects.19,20

Arterial blood pressure was measured by cuff sphygmomanometry at the time of the Doppler examination, and it was within the normal range in each of the patients and control subjects.

Echocardiography

M-mode echocardiograms recorded in patients with thalassemia and in normal controls were coded, and measurements were obtained by one observer who had no knowledge of the identity of the subjects. In each subject, measurements were obtained from four to six cardiac cycles and values were averaged. Ventricular septal and left ventricular posterior wall thickness, left ventricular end-diastolic and end-systolic cavity dimension, and left atrial cavity dimension were measured according to the recommendations of the American Society of Echocardiography.21 Left ventricular percentage of fractional shortening was calculated as internal end-diastolic dimension minus internal end-systolic dimension divided by internal end-diastolic dimension multiplied by 100.
Because echocardiographic measurements were obtained according to the recommendations of the American Society of Echocardiography, left ventricular mass was calculated using the following formula: Mass (g) = 0.80 \( [1.04 \times (\text{left ventricular internal end-diastolic dimension} + \text{ventricular septum thickness} + \text{posterior wall thickness})^3 - (\text{left ventricular internal end-diastolic dimension})^3] + 0.6 \). Left ventricular mass calculated by this formula has been shown to correlate closely with left ventricular mass calculated by the Penn convention method.22

**Statistical Analysis**

Data were expressed as mean±SD. Differences between continuous variables were determined by using the unpaired or paired Student’s t test as appropriate. Upper and lower 95% normal confidence limits for the Doppler diastolic indexes were calculated from the two tails of the Student’s t test distribution by using the following formulas: Mean + (2.042×SD), and mean – (2.042×SD), respectively.23 A p value of less than 0.05 was considered statistically significant.

**Results**

**Clinical Findings**

The hematologic profile of the 32 patients with thalassemia major is summarized in Table 1. In the patient group, hemoglobin levels at the time of cardiac evaluation were 13.3±0.9 g/dl, and hemoglobin was ≥11.7 g/dl in each patient. Heart rate was similar and not significantly different in patients and controls (73±11 and 74±12 beats/min, respectively). Both systolic and diastolic arterial pressure values were slightly but significantly lower.
in the patients with thalassemia (109±10 and 69±7 mm Hg) compared with controls (115±10 and 73±8 mm Hg, respectively; p<0.02). Although patients and control subjects were matched for age and gender, body surface area was significantly smaller in the patient group (1.38±0.27 m²) than in the controls (1.60±0.24 m²; p<0.001).

Doppler Echocardiographic Findings

Doppler echocardiographic findings in patients with thalassemia and in normal control subjects are summarized in Table 2. Left ventricular filling patterns differed substantially in the two groups. Peak flow velocity in early diastole (E) and rate of deceleration of flow velocity after the early diastolic peak (EF) were significantly increased (p<0.01 and p<0.001, respectively), and flow velocity dt was reduced (p<0.001) in patients with thalassemia compared with controls (Figure 2). The ratio between the early and late (atrial) peaks of flow velocity (E/A) was also significantly higher in patients than in controls (p<0.001) (Figure 2). This increase in flow velocity at mitral valve opening followed by an abrupt and premature decrease of flow velocity in early diastole is usually described as a "restrictive" Doppler pattern of left ventricular filling11-13 and has been shown to reflect a decrease in left ventricular compliance.24 Duration of isovolumic relaxation (A2-D) and peak flow velocity during atrial contraction (A) did not differ significantly in patients and controls. An example of a restrictive Doppler pattern of left ventricular filling in a patient with thalassemia is shown in Figure 1 (panel B).

When individual patient analysis was performed, one or more Doppler diastolic indexes were beyond the 95% confidence limits obtained from the normal controls in 16 (50%) of the 32 study patients (Table 2). The most common diastolic abnormalities were increased rate of deceleration of flow velocity in early diastole (11 patients), and increased ratio between early and late peaks of flow velocity (six patients).

To exclude the possibility that the abnormal filling pattern identified in our study patients could be due in part to changes in ventricular load after blood transfusion, we also measured the Doppler diastolic indexes immediately before and within an hour of transfusion in a subgroup of 20 study patients. Doppler measurements (obtained by an observer who was blind to the temporal succession of the recordings) were similar and not significantly different before and after transfusion (Table 3).

M-Mode Echocardiographic Findings

M-mode echocardiographic findings in patients with thalassemia and in normal controls are summarized in Table 4. By selection criteria, left ventricular end-diastolic cavity dimension and systolic fractional shortening in the study patients with thalassemia were within normal limits (<54 mm and ≥30%, respectively). Ventricular septal and posterior free wall thickness, and left atrial cavity dimension were similar and not significantly different in patients and controls. Calculated left ventricular mass and mass index were also similar in the two groups.

Relation Between Chelation Therapy and Diastolic Filling Patterns

To determine whether iron chelation with deferoxamine can protect from development of left ventricular diastolic abnormalities, Doppler diastolic indexes

| Table 1. Hematologic Profile of 32 Patients With Thalassemia Major |
|---------------------|------------------|------------------|-----------------|------------------|
|                     | Mean±SD          | Mean±SD          | Maximum serum†  | Total transfused |
|                     | (yr)             | (n) (units)      | ferritin (ng/ml)| iron burden (g) |
| Mean±SD             | 17±5             | 348±100          | 2,169±1,699     | 4,140±2,757      |
| Range               | 9–28             | 180–591          | 539–7,150       | 1,600–11,671     |

*Mean of three to five values obtained in each patient during the year before cardiac evaluation.
†Maximum value obtained in each patient since onset of transfusion therapy.

| Table 2. Doppler Diastolic Indexes in 32 Patients With Thalassemia Major and 32 Age-Matched and Sex-Matched Normal Control Subjects |
|---------------------|------------------|------------------|------------------|------------------|
|                     | Patients with    | Control subjects | p*               | 95% Confidence   |
|                     | thalassemia      |                  |                 | limits‡          |
| RR (msec)           | 822±119          | 815±133          | NS              | ……              |
| A2–D (msec)         | 44±10            | 47±10            | NS              | 27–67            |
| E (cm/sec)          | 90±10            | 81±15            | <0.01           | 50–112           |
| EF slope (cm/sec²)  | 1,050±325        | 763±193          | <0.001          | 369–1,157        |
| dt (msec)           | 97±22            | 119±19           | <0.001          | 80–158           |
| A (cm/sec)          | 35±7             | 39±10            | NS              | 19–59            |
| E/A                 | 2.7±0.7          | 2.2±0.5          | <0.001          | 1.0–3.2          |

A2-D, duration of isovolumic relaxation; E, peak early diastolic flow velocity; EF slope, rate of decrease of flow velocity in early diastole; dt, flow velocity deceleration time; A, peak late diastolic flow velocity; E/A, ratio of peak flow velocity in early diastole to peak flow velocity in late diastole.

*Derived from 32 age-matched and sex-matched normal control subjects.
TABLE 3. Doppler Diastolic Indexes Immediately Before and After Transfusion in 20 Patients With Thalassemia Major*

<table>
<thead>
<tr>
<th>Doppler indexes</th>
<th>Before transfusion</th>
<th>After transfusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>RR (msec)</td>
<td>771±131</td>
<td>793±123</td>
</tr>
<tr>
<td>Ar-D (msec)</td>
<td>44±9</td>
<td>41±9</td>
</tr>
<tr>
<td>E (cm/sec)</td>
<td>85±13</td>
<td>87±14</td>
</tr>
<tr>
<td>EF slope (cm/sec²)</td>
<td>1,112±256</td>
<td>1,060±298</td>
</tr>
<tr>
<td>dt (msec)</td>
<td>99±16</td>
<td>103±20</td>
</tr>
<tr>
<td>A (cm/sec)</td>
<td>35±9</td>
<td>33±7</td>
</tr>
<tr>
<td>E/A</td>
<td>2.7±1.2</td>
<td>2.8±1.2</td>
</tr>
</tbody>
</table>

Ar-D, duration of isovolumic relaxation; E, peak early diastolic flow velocity; EF slope, rate of decrease of flow velocity in early diastole; dt, flow velocity deceleration time; A, peak late diastolic flow velocity; E/A, ratio of peak flow velocity in early diastole to peak flow velocity in late diastole.

*None of the differences in values before and after transfusion were significant (paired Student's t test).

were analyzed in the subgroup of 16 patients who underwent optimal chelation therapy. Data obtained in these patients were compared with those obtained in 16 age-matched and sex-matched control subjects (Table 5). An abnormal pattern of left ventricular diastolic filling, with increased rate of deceleration of flow velocity after the early diastolic peak (EF) and reduced flow velocity dt, was also identified in this subgroup of patients who had undergone scrupulous chelation therapy. One or more Doppler diastolic indexes were beyond the 95% normal confidence limits in six (38%) of these 16 patients.

**Discussion**

Congestive heart failure due to iron overload of the myocardium is the most common cause of death in transfused patients with thalassemia major.1-2 In the advanced stages of the disease, cardiac abnormalities are characterized by systolic dysfunction, with left ventricular dilatation and reduced ejection fraction.1-4 In patients with normal systolic function at rest, a subnormal ejection fraction response to exercise has been identified by radionuclide angiography.25,26

Left ventricular diastolic filling has never been investigated systematically in patients with thalassemia major. A restriction to ventricular diastolic filling, however, has been described in some patients with idiopathic hemochromatosis.5-7 In the present study, we used Doppler echocardiography to assess left ventricular diastolic filling in a relatively large group of patients with thalassemia major who were free of cardiac symptoms and had normal left ventricular systolic function and diastolic cavity dimension. Our findings show that diastolic filling is altered in an early stage of this disease, when iron overload has not yet caused systolic dysfunction and left ventricular dilatation. Peak transmitral flow velocity, rate of deceleration of flow velocity in early diastole, and the ratio between the early and late (atrial) peaks of flow velocity were increased, and flow velocity deceleration time was reduced in our study patients with thalassemia compared with normal control subjects. These alterations in the Doppler transmitral waveform, with increased flow velocity at mitral valve opening followed by an abrupt decrease of flow velocity in early diastole, are usually described as restrictive11-13 and reflect an increase in left ventricular chamber stiffness.24 Conversely, duration of isovolumic relaxation was similar in patients and controls, a finding consistent with the concept that duration of relaxation can be normal in the presence of increased chamber stiffness.11-13,24 It should be pointed out that diastolic filling was not altered in all study patients; individual patient analysis identified a restrictive filling pattern in 50% of the patients. Nevertheless, the progressive nature of the cardiac complications associated with long-term iron overload suggests that alterations in diastolic filling will probably develop in all patients with thalassemia at some time during the clinical course of the disease.

TABLE 4. M-Mode Echocardiographic Measurements in 32 Patients With Thalassemia Major and in 32 Age-Matched and Sex-Matched Normal Control Subjects*

<table>
<thead>
<tr>
<th>Echocardiographic measurements</th>
<th>Patients with thalassemia</th>
<th>Control subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td>VS thickness (mm)</td>
<td>8±2</td>
<td>8±1</td>
</tr>
<tr>
<td>PW thickness (mm)</td>
<td>7±2</td>
<td>7±1</td>
</tr>
<tr>
<td>LVlDd (mm)</td>
<td>46±5</td>
<td>46±5</td>
</tr>
<tr>
<td>LVlDs (mm)</td>
<td>30±4</td>
<td>29±4</td>
</tr>
<tr>
<td>%FS</td>
<td>35±4</td>
<td>37±5</td>
</tr>
<tr>
<td>LA (mm)</td>
<td>35±5</td>
<td>32±4</td>
</tr>
<tr>
<td>Mass (g)</td>
<td>111±41</td>
<td>109±36</td>
</tr>
<tr>
<td>Mass index (g/m²)</td>
<td>78±18</td>
<td>67±15</td>
</tr>
</tbody>
</table>

*VS, ventricular septum; PW, posterior wall; LVlDd, left ventricular internal diastolic diameter; LVlDs, left ventricular internal systolic diameter; %FS, percentage of left ventricular fractional shortening; LA, left atrium.

*None of the differences in values were significant (unpaired Student's t test).

TABLE 5. Doppler Diastolic Indexes in 16 Patients With Thalassemia Major Who Underwent Optimal Chelation Therapy and in 16 Age-Matched and Sex-Matched Normal Control Subjects

<table>
<thead>
<tr>
<th>Doppler indexes</th>
<th>Chelated patients</th>
<th>Control subjects</th>
<th>p*</th>
</tr>
</thead>
<tbody>
<tr>
<td>RR (msec)</td>
<td>789±97</td>
<td>832±142</td>
<td>NS</td>
</tr>
<tr>
<td>Ar-D (msec)</td>
<td>48±9</td>
<td>45±10</td>
<td>NS</td>
</tr>
<tr>
<td>E (cm/sec)</td>
<td>88±8</td>
<td>83±14</td>
<td>NS</td>
</tr>
<tr>
<td>EF slope (cm/sec²)</td>
<td>1,080±356</td>
<td>795±214</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>dt (msec)</td>
<td>94±19</td>
<td>118±19</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>A (cm/sec)</td>
<td>37±6</td>
<td>39±9</td>
<td>NS</td>
</tr>
<tr>
<td>E/A</td>
<td>2.5±0.4</td>
<td>2.2±0.6</td>
<td>NS</td>
</tr>
</tbody>
</table>

Ar-D, duration of isovolumic relaxation; E, peak early diastolic flow velocity; EF slope, rate of decrease of flow velocity in early diastole; dt, flow velocity deceleration time; A, peak late diastolic flow velocity; E/A, ratio of peak flow velocity in early diastole to peak flow velocity in late diastole.

* Differences were determined by using the unpaired Student's t test.
The loading conditions of the ventricle can influence ventricular filling independently of the intrinsic diastolic properties of the myocardium.\textsuperscript{27,28} Therefore, to exclude the possibility that the abnormal filling pattern identified in our patient population could be due in part to changes in ventricular load after blood transfusion, we also assessed Doppler diastolic indexes immediately before and after transfusion in a subset of our study patients. The pattern of left ventricular filling, however, remained unchanged.

A previous echocardiographic investigation has emphasized that increased wall thickness and mass are characteristic of thalassemia major.\textsuperscript{4} More than 30% of the patients reported in this previous study, however, had an enlarged heart on routine chest roentgenogram, and many had symptoms of cardiac failure. Thus, these patients were in an advanced stage of the disease. Conversely, left ventricular wall thickness, mass, and mass index were normal in our study patients. This absence of gross morphological abnormalities reflects our patient selection criteria because our study population was confined to patients with a nondilated left ventricle and without symptoms of heart failure.

Since the late 1970s, iron chelation with deferoxamine has become a routine therapy in patients with thalassemia major.\textsuperscript{29–31} Preliminary observations in patients who began treatment in the second decade of life suggest that deferoxamine might protect from cardiac damage induced by iron overload.\textsuperscript{32,33} Children in whom routine chelation treatment was started shortly after the onset of transfusion therapy, however, are not yet at the end of their second decade of life, the age at which cardiac involvement usually becomes clinically manifest. Thus, the efficacy of long-term chelation treatment with deferoxamine is still uncertain. To determine whether iron chelation can prevent the development of diastolic filling abnormalities, we compared Doppler diastolic indexes obtained in a subgroup of our study patients who underwent optimal chelation treatment with those obtained in normal control subjects. A restriction to left ventricular filling was identified also in these patients despite their scrupulous chelation therapy.

It should be pointed out that Doppler diastolic indexes showed overlap in the overall group of patients with thalassemia and in normal subjects. Thus, the findings of the present study should be used cautiously to draw conclusions regarding cardiac damage in individual patients with this disease. It should also be emphasized that the prognostic implications of these findings remain to be clarified and need to be defined on the basis of prospective investigations.

The results of the present study demonstrate that left ventricular diastolic filling is altered in patients with thalassemia major and that diastolic abnormalities develop in a preclinical phase of cardiac involvement, when symptoms of heart failure are absent and systolic function is normal. Our findings also suggest that iron chelation therapy with deferoxamine might not completely protect patients with thalassemia from myocardial damage due to iron-related cardiac toxicity.

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KEY WORDS • thalassemia • hemochromatosis • diastolic function
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