Marrow Transplantation for Patients With Thalassemia: Results in Class 3 Patients

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Thalassemia patients can be categorized as class 1 (minimal liver damage and iron overload), class 3 (extensive liver damage from iron overload), and class 2 (intermediate). These categories are prognostic for treatment outcome after marrow transplantation. Class 3 patients have more transplant-related mortality than other patients. This study examines transplantation outcome for class 3 patients. Records were reviewed of 215 patients in class 3 who received transplants in Pesaro from HLA-identical related donors between May 1, 1984 and May 1, 1994. The influence of pretransplant, peritransplant, and posttransplant variables on survival, relapse, and transplant-related mortality was examined by product-limit and proportional-hazards multivariate analysis. Age and conditioning regimen were influential on survival, and regimens with less than 200 mg/kg cyclosporine (CY) were associated with 5-year survival probabilities of .74 and .63 in patients younger than 17 years and older patients, respectively. Transfusion history and regimen were influential on rejection, with 5-year probabilities of .53 and .24 in patients who received less than or greater than 100 red blood cell transfusions before transplantation and regimens containing less than 200 mg/kg CY. Results of transplantation for patients with advanced thalassemia treatment have improved with the introduction of conditioning regimens with less CY. This has been associated with an increase in rejection (particularly in patients who have received <100 red blood cell transfusions before transplant). Efforts at reducing the rejection rate by modifying the conditioning regimen should be concentrated on younger patients who have received a small number of transfusions. Patients with thalassemia who have HLA-identical family members should be transplanted before they are in class 3.

MARROW TRANSPLANTATION from HLA-identical related donors is an important option in the treatment of patients with homozygous β-thalassemia, and early transplantation is clearly the best option for some patients.[1]

Patient categories that are prognostically significant for the outcome of marrow transplantation have been described and tested in patients younger than 17 years of age.[2] Three classes were identified on the basis of adequate chelation as part of conventional therapy, hepatomegaly, and the presence of portal fibrosis in the liver. Results of transplantation in class 1 patients (who are all <17 years of age) have been described, and it was concluded that because of the high probability of cure with low early and late morbidity and mortality, there was no reason to deny class 1 patients with HLA-identical donors the advantages of a life free from tedious, expensive, and uncomfortable therapy.[3] Transplantation of such patients produced a 92% projected probability of long-term survival and an 85% probability of surviving disease-free for longer than 10 years. Results are marginally worse for children in class 2,[4] with long-term survival and disease-free survival of 84% and 80%, respectively, because of a small increase in nonrejection mortality as compared with class 1 patients.

We have described the results of transplantation of adults when the conditioning regimen was assigned on the basis of degree of thalassemia at the time of transplantation.[5] All patients were class 2 or 3, and marrow transplantation was undertaken because disease progression was occurring despite conventional therapy. Probabilities of survival, disease-free survival, and rejection were .85, .80, and .05, respectively, with a survival plateau extending from 6 months to 3 years, and it was concluded that marrow transplantation is effective therapy that should be offered to adults with progressive thalassemia who have suitable donors.[6]

Early results indicated that children in class 3 had a much worse outcome, with probabilities of survival, disease-free survival, and rejection of .61, .53, and .16, respectively. This was largely due to a major increase in nonrejection mortality.[2,7] Marrow transplantation of such patients has been the subject of intensive study, because the clear deterioration of such patients while undergoing transfusion/chelation therapy has provided a major incentive for intervention.

We now report the Pesaro experience since May 1984 in transplanting patients with class 3 thalassemia.

SUBJECTS AND METHODS

Patient categorization. A system has been described for assigning patients undergoing marrow transplantation for thalassemia to prognostically useful categories.[2] Risk factors evaluated were hepatomegaly (>2 cm below the intercostal margin), presence of portal fibrosis in the pretransplant liver biopsy, and quality of chelation during the years before transplant. The quality of chelation was characterized as regular when deferoxamine therapy was initiated within 18 months of the first transfusion and administered subcutaneously for 8 to 10 hours continuously for at least 5 days each week. The chelation variable was defined as irregular for any deviation from this requirement. The age in months when the patient first received regular chelation was recorded. A chelation index was calculated describing the number of months each patient received regular chelation as a percentage of the number of months the patient should have received chelation by the definition above. With this index, a completely satisfactory chelation history is represented by 100% and completely unsatisfactory by 0%. Fifty-nine of 151 pa-

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Submitted March 27, 1995; accepted October 5, 1995.

Supported by the Italian Association Against Cancer, Milano, the Italian Association Against Leukemia, Pesaro, the Berloni Foundation Against Thalassemia, Pesaro, and the Regione Marche, Ancona.

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0006-4971/96/8705-0014$3.00/0

tients younger than 17 years and 14 of 63 patients aged 17 years or older did not receive satisfactory chelation for any period before transplantation. The time spent receiving transfusions without chelation, sometimes the largest part of the life of these patients, is a marker for the chronic organ damage incurred during periods with high iron overload.

Some of these patients had relatively low ferritin levels at the time of transplant, because they had received intensive chelation during the months before transplant. We believe that organ damage induced by periods of inadequate chelation is not reversed by a subsequent period of intensive chelation, although this may decrease ferritin levels. Our experience with patients who have had successful transplants is that it takes a very long time to reverse iron overload–associated organ damage, even in patients whose iron metabolism is not impacted by continuing thalassemia. We do not use ferritin levels in categorizing transplantation risk, and our analyses confirm that they are not independently predictive when chelation history (which is univariately of greater significance) is entered into the statistical model.[2] We believe that organ damage acquired during periods when transfusions were not covered by deferoxamine persists and is higher than may be indicated by measurements of iron overload at the time of transplant, particularly if adequate chelation has been given in recent years.

Class 1 patients had no adverse risk factors, class 3 patients had all three, and class 2 patients had one or two adverse risk factors. All patients in this report were class 3 by the above categorization.

Patient characteristics. Informed consent was obtained from the parents of all minor patients after the procedures and risks involved in marrow transplantation were explained in detail. Explanations to the patients themselves varied in complexity according to age. Particular emphasis was placed on the option of continued conventional management with transfusions and deferoxamine.

From May 1, 1984 to May 1, 1994, 215 patients categorized as class 3 received marrow transplants in Pesaro from HLA-identical related donors. One patient was prepared for transplantation with a customized regimen designed to accommodate particular pretransplant problems and has been excluded from analysis. The donors were genotypically identical siblings of the recipients in 206 cases and phenotypically HLA-identical parents in eight cases (six fathers and two mothers).

All patients had received red blood cell transfusions before transplantation. In two cases, the first red blood cell transfusion was delayed for a very long period, with one patient having the first transfusion at 13 years and the other at 22 years of age. Both of these patients had homozygous β°-thalassemia with intermedia expression of the clinical disease.

Characteristics potentially influential on the outcome of transplantation are presented in Tables 1 and 2.

Liver biopsies were obtained from all patients in this series. Recently, we published an assessment of the reliability and safety of percutaneous liver biopsy in evaluating hepatic iron loading and histology. In this study, the fibrosis score of multiple samples of liver obtained at autopsy within 100 days of percutaneous biopsy in 41 patients who died following marrow transplantation was clearly correlated with the first sample in greater than 60% of cases. In most of the discordant cases, fibrosis had been underestimated, so it is unlikely that a class 2 patient was erroneously assigned to class 3.[8] The grading systems used to evaluate siderosis, chronic active hepatitis, chronic persistent hepatitis, and portal fibrosis have been reported.[9] Three grades of severity (mild, moderate, and severe) were identified for each diagnostic category. The histopathology of the liver at the time of transplant is described in Table 2. All patients had portal fibrosis and siderosis, 34% had chronic persistent hepatitis, and 23% had chronic active hepatitis.

Positive serology for exposure to hepatitis B was present in 126 patients (59%). Second-generation tests for a history of hepatitis C virus exposure were available for 71 patients younger than 17 years and 47 patients older than 16 years.

Table 1. Patient Characteristics (N = 214)

Characteristic	No. of Patients	Range	Median	Mean ± SD
Age, (yr)				
<17	151	3-16	11.0	11 ± 3.2
≥17	63	17-32	19	20.4 ± 3.3
Gender				
Male	130			
Female	84			
No. of red blood cell transfusions				
<17 yr	151	4-435	150	158.0 ± 87.2
≥17 yr	63	130-600	300	332.7 ± 122.8
Months of age at first transfusion				
<17 yr	151	2-156	8	15.3 ± 20.3
≥17 yr	63	3-264	10	23.1 ± 37.5
Months of age at first chelation				
<17 yr	93	24-180	60	66.8 ± 29.6
≥17 yr	49	36-240	90	98.7 ± 41.4
Chelation index*				
<17 yr	92 (>0%)	10-98	74	68 ± 22.1
≥17 yr	49 (>0%)	25-97	72	71 ± 16.5
Serum ferritin (ng/mL)				
<17 yr	151	604-17,450	3,374	4,025 ± 2,631
≥17 yr	63	328-9,071	1,911	2,653 ± 2,014
Serum bilirubin (mg/dL)				
<17 yr	151	0.1-7	1.1	1.3 ± 0.98
≥17 yr	63	0.2-5.9	1.2	1.6 ± 1.15
Aspartate aminotransferase (IU/L)				
<17 yr	151	5-406	34	50.7 ± 59.6
≥17 yr	63	11-225	30	45.5 ± 41.9
Alanine aminotransferase (IU/L)				
<17 yr	151	6-742	62	88.7 ± 106.4
≥17 yr	63	12-353	52	72.1 ± 65.6
Hepatitis B seropositive (positive/tested)				
<17 yr	83/151			
≥17 yr	43/63			
Hepatitis C seropositive (positive/tested)				
<17 yr	33/71			
≥17 yr	41/47			

* Months chelation was actually given as a percent of months when it was indicated.

Transplant regimens. All the conditioning regimens used busulfan (BU) and cyclophosphamide (CY), and some regimens included antilymphocyte globulin (Lymphoglobuline; Merieux, Paris, France) for additional immunosuppression. Table 3 presents the distribution of various conditioning regimens and graft-versus-host disease (GVHD) prophylactic regimens by age.

BU 14 or 16 mg/kg was administered orally three times daily in

Table 2. Pretransplant Liver Histopathology

Characteristic	No. of Subjects	Severity				
		NO	MI	MO	SE	CI
Portal fibrosis						
<17 yr	151	0	36	50	60	5
≥17 yr	63	0	18	14	26	5
Siderosis						
<17 yr	151	0	11	56	84	—
≥17 yr	63	0	12	27	24	—
Chronic persistent hepatitis						
<17 yr	150*	103	46	1	0	—
≥17 yr	63	36	26	1	0	—
Chronic active hepatitis						
<17 yr	150	85	36	26	3	—
≥17 yr	63	39	12	10	2	—

Abbreviations: NO, none; MI, mild; MO, moderate; SE, severe; CI, cirrhotic.

* Data on hepatitis histopathology were not available for one patient <17 years of age.

14 doses over 4 days, followed by intravenous CY 50 or 40 mg/kg daily each of the next 4 days or 60 mg/kg each of the next 2 days. Antilymphocyte globulin was administered at a dose of 10 mg/kg intravenously. Marrow was infused 36 hours after the last dose of CY. The date of marrow infusion was designated day 0. All patients received 7.5 mg/kg CY intravenously on day 1. Methotrexate (MTX) was administered as 10 mg/m^2 intravenously. Cyclosporine (CSP) was administered as 5 mg/kg daily intravenously from day −3 through day 5, and then reduced to 3 mg/kg/d intravenously until oral administration of 12.5 mg/kg/d could be tolerated.[10,11] The dose of CSP was tapered from day 60 until discontinuation at 1 year. Antilymphocyte Globuline was administered at a dose of 10 mg/kg intravenously.

Supportive measures. All patients were treated in positive-pressure isolation rooms and received oral nonabsorbable antibiotics and a low-bacteria diet. Systemic prophylaxis against infection was achieved with amikacin and piperacillin or amikacin and ceftazidime, which started on the day before transplantation and continued until the granulocyte count exceeded 0.5 × 10^9/L and the patient was afebrile, together with intravenous amphotericin B from day 8 until discharge from hospital. All blood products administered after transplantation were irradiated with 30 Gy. Acute and chronic GVHDs were graded according to the Seattle criteria.[12,13] The first-choice drug for treatment of acute GVHD was prednisolone at escalating doses up to 10 mg/kg/d depending on the degree and duration of GVHD. The criteria used for the diagnosis of veno-occlusive disease were previously described by Jones et al.[14]

Tests of chimerism. Cytogenetic analyses were performed on unstimulated marrow and phytohemagglutinin-stimulated peripheral blood of both donor and recipient if they were of opposite sex. Globin-chain synthesis of marrow and peripheral blood reticulocytes was examined via incorporation of [^{3}H]-leucine followed by column or high-performance liquid chromatography. Analyses of karyotype and of globin synthesis pattern were performed on peripheral blood and bone marrow on day 13 and repeated at 2-week intervals until day 60. Jeffreys minisatellite single-locus DNA probes (MS1, MS31, MS43, and g3) were used to distinguish donor and recipient cells in restriction fragment length polymorphism analysis of variable number tandem repeat loci.[15,16]

Statistical analysis. Survival distributions were estimated by the product-limit method,[17] tested for equality by the Mantel-Cox[18] and Breslow[19] statistics. In estimating event-free survival, we identified rejection, recurrence of thalassemia, and death as events. Rejection was defined as development of complete marrow aplasia or recurrence of thalassemia (a return to the pretransplant pattern of globin-chain synthesis).

Cox proportional-hazards analysis was used in a stepwise model to evaluate the influence of pretransplant variables on survival and rejection-free survival. Variables examined were patient and donor gender and their permutations, patient age, transfusion history, transplant regimen, degree of portal fibrosis, interval to first transfusion and to initiation of chelation therapy, quality of chelation therapy, serum ferritin, presence of severe or moderate chronic active hepatitis, elevated serum bilirubin, and elevated transaminases.

Long-term follow-up study. Patients were advised to return to Pesaro for evaluation at 6 months after transplantation, again at 1 year, and annually thereafter. The marrow and peripheral blood were examined by chromatography and, when appropriate, cytogenetics at the 6-month, 1-year, and 2-year visits. Other characteristics evaluated included iron metabolism, liver, cardiac, and endocrine function, and growth and development.

RESULTS

Figure 1 presents Kaplan-Meier probabilities of survival, event-free survival, rejection, and nonrejection mortality for 214 patients in this study.

The first conditioning regimen used for these class 3 patients consisted of 14 mg/kg BU and 200 mg/kg CY. Among the early patients treated with this regimen, five were older than 16 years of age, four of whom died of transplant-related

Table 3. Conditioning Regimens and GVHD Prophylaxis

Conditioning Regimen				No. of Patients	
BU/CY	Antilymphocyte Globuline	MTX*	Cyclosporine†	<17 yr	>16 yr
14/200	No	Days 3, 6, and 11, and weekly for 100 days	No	2	1
14/200	No	No	1 yr	44	2
14/200	No	Days 3, 6, and 11	1 yr	10	2
14/120	day −5 to +5	Days 3, 6, and 11	1 yr	19	15
14/120	day −5 to +5	No	1 yr	8	0
16/120	day −5 to +5	Days 3, 6, and 11	1 yr	7	1
16/120	No	Days 3, 6, and 11	1 yr	28	13
16/160	No	Days 3, 6, and 11	1 yr	21	18
14/160	day −7 to −2	Days 3, 6, and 11	1 yr	12	11

* Administered as 10 mg/m^2 intravenously.

† Administered as 5 mg/kg daily intravenously from day −3 through day 5, and then reduced to 3 mg/kg/d intravenously until oral administration of 12.5 mg/kg/d could be tolerated.[10,11]

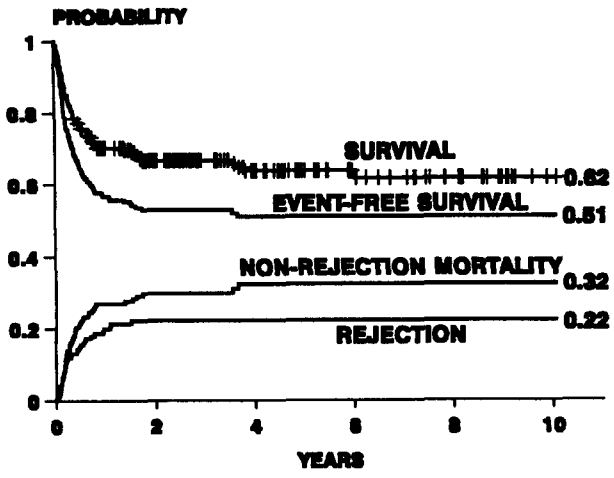

Fig 1. Kaplan-Meier probabilities of survival, rejection-free survival, nonrejection mortality, and rejection for 214 patients transplanted from HLA-identical related donors for treatment of class 3 homozygous β°-thalassemia.

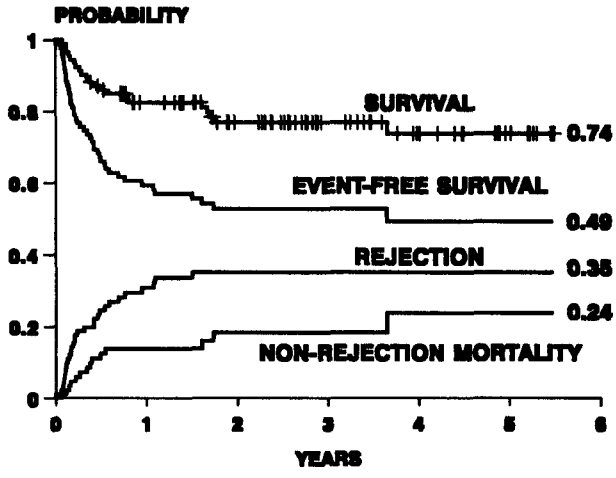

Fig 3. Kaplan-Meier probabilities of survival, rejection-free survival, nonrejection mortality, and rejection for 95 patients aged <17 years transplanted from HLA-identical related donors for treatment of class 3 homozygous β°-thalassemia with conditioning regimens containing <200 mg/kg CY.

causes soon after transplant, and one, who rejected the transplant, died at 6 years after transplant of causes related to recurrent thalassemia. Because of this early experience, attempts at marrow transplantation of patients older than 16 years were abandoned while efforts were made to improve the results of transplanting class 3 patients. Thus, there were 61 patients who received this conditioning regimen, and 56 of these were aged less than 17 years. Figure 2 presents Kaplan-Meier statistics for these patients.

Subsequent patients less than 17 years of age were treated with conditioning regimens containing less than 200 mg/kg CY. Figure 3 presents survival, event-free survival, rejection, and nonrejection mortality for 95 children treated with these regimens. Treatment of patients aged 17 years or older was resumed using the revised regimens, and Fig 4 shows Kaplan-Meier statistics for 58 adult class 3 patients so treated.

Of 153 patients receiving regimens with less than 200 mg/kg CY, 73 received antilymphocyte globulin, and 73 of these were younger than 17 years. The use of antilymphocyte globulin had no detectable influence on probabilities of survival or of relapse in these patients.

Multivariate analyses. After stepwise multivariate analyses of the effect of the variables listed above on survival for the entire group of 214 patients, only age less than 17 years emerged as independently beneficial. Regimens containing 200 mg/kg CY (v <200 mg/kg), presence of severe portal fibrosis, and serum bilirubin greater than 2.0 mg/dL were independently adversely influential on survival (Table

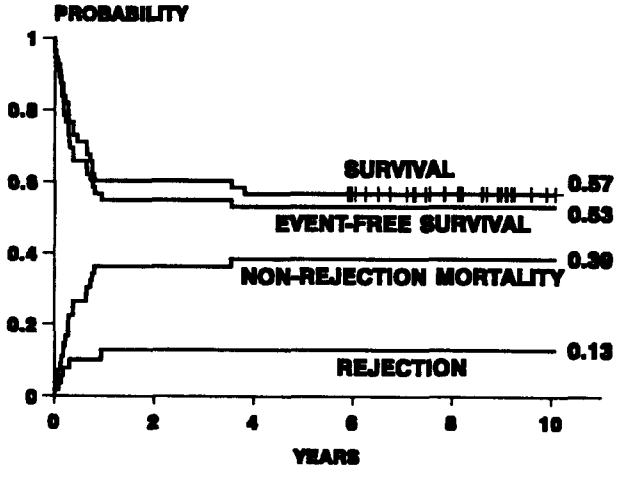

Fig 2. Kaplan-Meier probabilities of survival, rejection-free survival, nonrejection mortality, and rejection for 56 patients aged <17 years transplanted from HLA-identical related donors for treatment of class 3 homozygous β°-thalassemia using a conditioning regimen consisting of 14 mg/kg BU and 200 mg/kg CY.

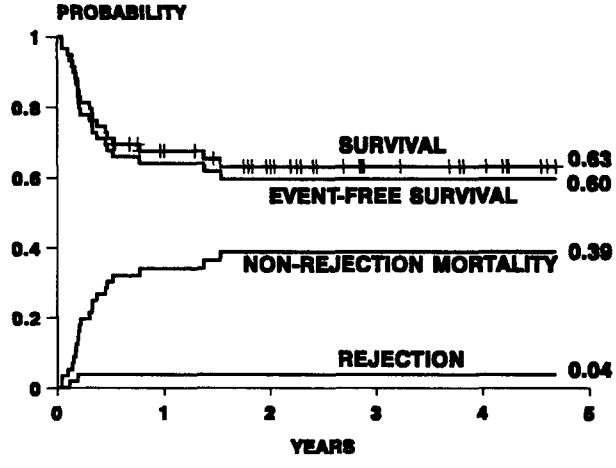

Fig 4. Kaplan-Meier probabilities of survival, rejection-free survival, nonrejection mortality, and rejection for 58 patients aged ≥17 years transplanted from HLA-identical related donors for treatment of class 3 homozygous β°-thalassemia with conditioning regimens containing <200 mg/kg CY.

Table 4. Cox Multivariate Analysis of Survival for 214 Class 3 Patients

Variable	P	Risk Ratio	95% Confidence Limits
Age <17 yr *v* older	.0075	0.498	0.29-0.85
Regimens with CY 200 mg/kg *v* <200 mg/kg	.0054	2.520	1.48-4.29
Severe *v* moderate or mild portal fibrosis	.0117	1.855	1.14-3.02
Serum bilirubin >2.0 mg/dL *v* <2.0 mg/dL	.0093	2.468	1.40-4.34
Severe acute GVHD	.0001	7.933	4.63-13.59

Entry of acute GVHD grade 3 or 4 removed the influence of age <17 years.

Table 5. Cox Multivariate Analysis of Rejection for 151 Class 3 Patients Less Than 17 Years Old

Variable	P	Risk Ratio	95% Confidence Limits
<100 transfusions *v* >100	.005	2.65	1.37-5.13
Regimens with CY 200 mg/kg *v* <200 mg/kg	.014	0.30	0.13-0.73
Aspartate aminotransferase >50 IU/L	.025	2.24	1.09-4.60

4). When the analysis was repeated with the addition of time-dependent covariates representing moderate or severe acute GVHD, severe acute GVHD emerged as independently adversely influential and age less than 17 years ceased to be an advantage, whereas the other variables were unaffected.

In multivariate analyses of rejection for the entire group of 214 patients, only transfusion history emerged as independently influential on the probability of rejection. Patients who received less than 100 red blood cell transfusions before transplantation had a risk ratio of 3.189 for rejection (P = .0001) as compared with patients who received more red blood cell transfusions (95% confidence limits, 1.71 and 5.93). Acute GVHD had no influence on the probability of rejection.

These analyses were repeated examining the population of patients aged less than 17 years. The only factor influential on survival was the conditioning regimen, and regimens containing 200 mg/kg were associated with a risk ratio of 2.04 (P = .016, confidence limits = 1.13 and 3.68). When acute GVHD was entered into the analysis, no other variable was independently significant and the risk ratio associated with severe acute GVHD was 7.04 (P = .0001, confidence limits = 3.51 and 14.12). In the population of patients less than 17 years of age, only pretransplant transfusion history, regimen, and elevated aspartate aminotransferase before transplant were independently associated with the probability of rejection (Table 5).

Patients who had received less than 100 red blood cell transfusions before transplant were significantly more likely to reject, whereas patients who received conditioning that included CY 200 mg/kg were significantly less likely to reject. The Kaplan-Meier probability of rejection was examined for patients less than 17 years of age who were conditioned with regimens that contained less than 200 mg/kg CY. For patients who had received less than 100 red blood cell transfusions, this probability was .53, and for those who had received more red blood cell transfusions, it was .24 (P = .003; Fig 5).

GVHD. The probability of developing moderately severe acute GVHD (grades 2, 3, or 4) was .15 for patients aged less than 17 years and .19 for patients aged 17 years or more. Of 151 patients aged less than 17 years, six developed moderate or severe chronic GVHD, and three of these died (two from infection associated with GVHD and one from AIDS contracted before transplantation). Two of these patients survive with complete resolution of chronic GVHD, and one has residual GVHD with a Karnofsky score of 80. Of 63 patients aged 17 years or older, three developed moderate or severe chronic GVHD, and two of these died (one from mushroom poisoning and one from infection associated with GVHD) and one survives with persistent chronic GVHD and a Karnofsky score of 80.

Causes of death. There were 13 deaths related to either failure to engraft, rejection, or disease recurrence. One patient died on day 37 from fungus infection without engraftment or recurrence. Two patients did not engraft, developed recurrent thalassemia, and died on days 77 and 294 from liver toxicity and infection, respectively. Seven engrafted patients died after recurrence of thalassemia. One of these deaths occurred at 42 days after transplant from cerebral hemorrhage, one died from a B-cell lymphoma on day 165, and the others died of causes related to recurrence of thalassemia. Three patients with initial engraftment rejected the grafts without thalassemic recurrence on days 21, 207, and 392, and all three died of infection.

A worrisome complication and cause of death after bone marrow transplantation has been sudden cardiac tamponade, which occurred in four cases (6%), most likely related to a particular susceptibility of the pericardial membrane of these patients.[20,21]

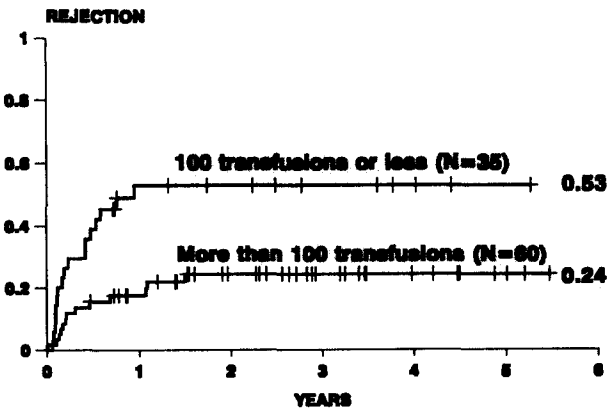

Fig 5. Kaplan-Meier probabilities of rejection for 95 patients aged <17 years transplanted from HLA-identical related donors for treatment of class 3 homozygous β^0-thalassemia with conditioning regimens containing <200 mg/kg CY. Patients are categorized on the basis of number of red blood cell transfusions received before transplantation.

Table 6. Causes of Death

Cause of Death	No.	%
Infection	33	46
AIDS	2	3
ARDS	1	1
IP	2	3
Acute GVHD	10	14
Cardiac tamponade	4	6
Hemorrhage	4	6
B-cell lymphoma	2	3
Venocclusive disease	1	1
Other liver failure	7	10
Died at home (unknown cause)	4	4
Mushroom poisoning	1	1

Table 6 presents the causes of all deaths in this series of patients.

DISCUSSION

Studies suggested that the mortality associated with being in the worst prognostic class for thalassemia (class 3) could be reduced by using regimens containing less CY,[7] and conditioning regimens for adults (patients ≥ 17 years of age) were thereafter selected on the basis of risk category. Only patients younger than 17 have been found to be in class 1, and we have reported our experience in treating such patients,[3] concluding that patients with thalassemia who have suitable donors should be treated by transplantation as soon as possible, because the mortality in class 1 patients was very low and the prospects for cure were high. Moreover, delay in transplantation presented the risk that patients would develop complications of iron overload, which would result in a less successful outcome for marrow transplantation.

The current report describes our total experience in transplanting patients with advanced liver damage resulting from thalassemia and its therapy (class 3 patients). Conditioning regimens with less than 200 mg/kg CY improved the Kaplan-Meier probability of survival for patients less than 17 years of age from .57 to .74, but were associated with an increase of rejection probability from .13 to .35. As a result of this increase in rejection rate, there was no improvement in rejection-free survival. The Kaplan-Meier probability of survival for patients older than 16 years who received conditioning regimens with less than 200 mg/kg CY was .63, but the rejection probability was not increased from that seen with the higher dose of CY, and consequently, rejection-free survival was better in the older group of patients.

Multivariate analysis of the outcome for all patients indicated that age, regimen, severe portal fibrosis, and elevated serum bilirubin levels influenced survival and only previous transfusion history influenced rejection, with patients who received less than 100 transfusions before transplant having a much higher probability of rejection. None of the patients older than 16 years had received less than 100 transfusions before transplantation. When the multivariate analysis was repeated examining only patients less than 17 years of age, only the amount of CY in the conditioning regimen was influential on survival and only the pretransplant transfusion history and aspartate aminotransferase were influential on rejection. The aspartate aminotransferase covariate had confidence limits of 1.09 to 4.60 for the risk ratio of 2.24, and considering the relatively large number of variables examined in this analysis, it is likely that the P of .03 is not biologically significant. There was no difference in survival or rejection probabilities associated with the use of CY doses of 160 versus 120 mg/kg.

Thus, age and regimen were important for survival, and regimens with less than 200 mg/kg CY were associated with survival probabilities of .74 and .63 in younger and older patients. Transfusion history and regimen were important for rejection, with rejection probabilities of .53 and .24 in patients who received less than or greater than 100 red blood cell transfusions before transplantation and regimens containing less than 200 mg/kg CY. Efforts at reducing the probability of rejection should therefore be directed at patients who have received less than 100 red blood cell transfusions before transplantation, although it is important to note that younger patients receiving more than this number will still have a substantial probability of rejection. Attempts to find a level of transfusion experience that is more informative about the prospects of rejection have been unsuccessful. It is not clear whether older patients intrinsically have a lower probability of rejection, since this probability was low in this group, but none had received less than 100 transfusions.

The lower rejection rate for patients who had received more transfusions was unexpected. In clinical and experimental marrow transplantation, an increase in the number of transfusions before transplantation has usually been associated with an increase in the probability of rejection, although the reverse effect has been seen in kidney transplantation. We have no information to support speculation about the cause of this effect in patients reported here.

The improvement in the results of transplantation of patients in class 3 is gratifying, but we must emphasize that the results are still very much worse than for transplantation in class 1. Moreover, we have evidence that the reversal of organ damage present before transplantation because of thalassemia and its treatment is much slower in class 3 patients than in class 1 patients.[22,23] We agree with the statement in a recent report[24] that patients with large iron stores after chelation therapy should be transplanted. However, a more constructive approach would be to transplant patients before they have deteriorated to this stage. We would prefer never to be confronted with patients in class 3 in need of transplantation, because we believe that those who have suitable donors should have been transplanted earlier.

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