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Hematopoietic Cell Transplantation as Curative Therapy for Idiopathic Myelofibrosis, Advanced Polycythemia Vera, and Essential Thrombocythemia

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ABSTRACT

A total of 104 patients, aged 18 to 70 years, with a diagnosis of chronic idiopathic myelofibrosis (CIMF), polycythemia vera (PV), or essential thrombocythemia (ET) with marrow fibrosis were transplanted from allogeneic (56 related and 45 unrelated) or syngeneic (n = 3) donors. Busulfan (BU) or total body irradiation (TBI)-based myeloablative conditioning regimens were used in 95 patients, and a nonmyeloablative regimen of fludarabine plus TBI was used in 9 patients. The source of stem cells was bone marrow in 43 patients and peripheral blood in 61 patients. A total of 63 patients were alive at a follow-up of 1.3–15.2 years (median, 5.3 years), for an estimated 7-year actuarial survival rate of 61%. Eleven patients had recurrent/persistent disease, of whom 8 died. Nonrelapse mortality was 34% at 5 years. Patients conditioned with targeted BU (plasma levels 800–900 ng/mL) plus cyclophosphamide (tBUCY) had a higher probability of survival (68%) than other patients. Dupriez score, platelet count, patient age, and comorbidity score were statistically significantly associated with mortality in univariate models. In a multivariable regression model, use of tBUCY ($P = .03$), high platelet count at transplantation ($P = .01$ for PV/ET; $P = .39$ for other diagnoses), younger patient age ($P = .04$), and decreased comorbidity score ($P = .03$) remained statistically significant for improved survival. Our findings show that hematopoietic cell transplantation offers potentially curative treatment for patients with ICMF, PV, or ET.

KEY WORDS

Myelofibrosis • Hematopoietic cell transplantation • Polycythemia vera • Essential thrombocythemia

INTRODUCTION

Chronic idiopathic myelofibrosis (CIMF), also known as agnogenic myeloid metaplasia, is a clonal chronic myeloproliferative disease characterized by progressive marrow fibrosis, extramedullary hematopoiesis with splenomegaly, anemia, and leukoerythroblastosis [1]. The median survival after diagnosis of CIMF is 3.5–5 years. Clinical presentations are heterogeneous. Prognostic factors include anemia, leukocytosis or leukopenia, clonal cytogenetic abnormalities, and older age. According to the Dupriez scoring system, which uses hemoglobin of < 100 g/L and white blood cell (WBC) counts $< 4 \times 10^9$/L or $> 30 \times 10^9$/L as risk factors, patients are divided into 3 risk groups: low risk (no risk factors), with a median survival of 7–8 years; intermediate risk (1 risk factor), with a median survival of 2.2 years; and high risk (2 risk factors), with a median survival of slightly more than 1 year [2]. This classification, although developed for conservatively managed patients, has also provided prognostic information for allogeneic hematopoietic cell transplantation (HCT). We and others have shown that, depending on Dupriez risk group, 40%–

80% of patients survive in remission after HCT [3-7]. Analysis of outcome data after HCT also suggests that in addition to hemoglobin and WBC, low platelet counts are significantly correlated with a lower probability of survival [5]. This observation agrees with a recent report from the Mayo Clinic showing that including platelet counts into the risk assessment of patients with CIMF improves prognostic accuracy in patients not undergoing transplantation [8]. Because HCT is currently the only therapy with curative potential for CIMF, these data suggest that patients with CIMF who are candidates for transplantation should be considered for transplantation before they develop high-risk features.

Myelofibrosis may also occur in patients with other myeloproliferative disorders, particularly polycythemia vera (PV) and essential thrombocythemia (ET). Although the overall prognosis for patients with PV or ET is much better than that for patients with CIMF, once myelofibrosis develops, no currently available treatment other than HCT has curative potential. Thus, it is noteworthy that the most encouraging transplantation results in patients with myelofibrosis have been achieved in those with a previous diagnosis of PV or ET [9], although because of transplantation-related mortality, the candidacy for and timing of HCT have remained controversial.

Here we provide a summary of transplantation results from a single center in 104 patients with myelofibrosis.

PATIENTS AND METHODS

Patient and Disease Characteristics

A total of 104 patients (57 females, 47 males) with a diagnosis of CIMF, post-PV or post-ET fibrosis, or myelofibrosis associated with myeloproliferative disorders not otherwise specified underwent HCT at the Fred Hutchinson Cancer Research Center (FHCRC). Fifty-four of these patients have been described previously [5]. Patient and disease characteristics are summarized in Table 1. All but 3 patients underwent transplantation after 1990. The patients ranged in age from 18 to 70 years (median, 49 years). The interval from diagnosis to transplantation ranged from 2 to 313 months (median, 15 months). The distribution by Dupriez risk group and severity of bone marrow fibrosis were similar to that in the original cohort of 54 patients; however, only 20% of patients in the new cohort had splenectomy, compared with 38% of the initial 54 patients. Also, the median patient age was 44 years in the original cohort but 53 years in the new cohort (51.2 years when excluding 9 patients conditioned with a nonmyeloablative regimen; see the following).

Indications for HCT included peripheral blood cytopenias, defined as a hemoglobin < 10 g/dL, platelet count $< 100 \times 10^9$/L, neutrophil count $< 1.5 \times 10^9$, or evidence of leukemic transformation. In 5 patients, myelofibrosis was possibly associated with another disease. One patient presented with bcr/abl-negative CIMF but 18 months later developed evidence of bcr/abl-positive CML; 1 patient was diagnosed concurrently with non-Hodgkin lymphoma; 2 patients exhibited some dysplastic marrow morphology, including megakaryocytes, in the context of extensive marrow fibrosis; and 1 patient had concurrent idiopathic thrombocytopenic purpura (ITP). Clonal karyotypic abnormalities were documented in 45 patients: +8 (± other abnormalities) in 8 patients, 20q− (± other abnormalities) in 6 patients, 7q− (± other abnormalities) in 2 patients, 13q− (± other abnormalities) in 2 patients, various translocations in 13 patients, and other structural or numeric abnormalities in 14 patients. Karyotypes were normal in 53 patients, and in 6 patients, material was insufficient for analysis. A total of 49 patients received transfusions of red blood cells, platelets, or both; 43 received hydroxyurea, 20 received interferon, 16 received anagrelide, 15

Table 1. *Patient and Disease Characteristics*

Data	Number of Patients
Number of patients studied	104
Patient age, (years), range (median)	18-70(49)
Patient sex, M/F	57/47
Disease duration, (months), range (median)	2-31(15)
Primary diagnosis	
CIMF	62
ET with myelofibrosis	18
PV with myelofibrosis	12
Myelofibrosis with increased blasts	7
Other*	5
Degree of marrow fibrosis†	
1	9
2	25
3	28
4	37
Dupriez risk classification at transplantation	
Low	44
Intermediate	40
High	20
Platelet count at transplantation	
$> 100 \times 10^9$/L	70
$< 100 \times 10^9$/L	34
Previous splenectomy‡	
Yes/no	29/69

*One patient had CIMF that evolved to CML. One patient had a concurrent diagnosis of non-Hodgkin's lymphoma, and 2 patients showed myelodysplastic changes in addition to the typical morphology of CIMF. One patient had idiopathic thrombocytopenic purpura in addition to marrow fibrosis.

†The degree of fibrosis was graded as described previously [10]. Five patients were not classified.

‡Data were incomplete for 6 patients.

received corticosteroids, 26 received erythropoietin, and 19 received chemotherapy other than hydroxyurea (30 patients received more than 1 agent); 12 received no therapy. Clinical, hematologic, and pathologic disease characteristics were assessed by standard procedures as described previously [4,5]. Marrow fibrosis was determined and staged as described previously [5,10]. All patients had given informed consent according to the requirements of the FHCRC's Institutional Review Board, which conforms to the Helsinki Declaration.

Donor Selection

Donor characteristics are summarized in Table 2. Donors ranged in age from 6 to 63 years (median, 41 years); 52 were male and 52 female. Fifty-nine patients had related donors: HLA genotypically identical siblings in 52, syngeneic twins in 3, and HLA-nonidentical family members in 4. Among the latter group, 1 differed for an HLA-DR antigen, 1 differed for -DR and -DQ antigens, 1 differed for a -B antigen (in the host-versus-graft direction), and 1 differed for -C and -B antigens (in the GVH direction). Unrelated donors were identified for 45 patients. High-resolution HLA-A, -B, -C, -DRB1, and -DQB1 allele typing was available in 34 patients, and 27 of these were HLA-identical with the recipients. Among the 18 donors who were not identical with the patients or had not undergone high-resolution typing, 9 were matched with their recipients by low- or intermediate-level typing. Another 7 were HLA-nonidentical by high-resolution typing (HLA-A locus in 3, -B locus in 2, -DQ locus in 1, and -DR plus -DQ loci in 1). The HLA data were incomplete in 2 donor–recipient pairs, and thus the degree of match or mismatch could not be determined in these 2 pairs.

Table 2. *Donor and Transplant Characteristics*

Data	Number of Transplants
Donor age (years), range (median)	**6-63 (41)**
Donor sex, M/F	**52/52**
Relationship	
Related	**59**
HLA-identical sibling	**52**
HLA-nonidentical	**4**
Syngeneic	**3**
Unrelated*	**45**
HLA-identical	**36**
HLA-nonidentical	**9**
Source of stem cells	
Marrow	**43**
Peripheral blood	**61**
Conditioning regimens	
BUCY/tBUCY	**6/59**
ATGtBUCY§	**14**
BUTBI (1200)	**9**
CYTBI (1200)	**6**
FLUtBU	**1**
FLUTBI (200)	**9**
GVHD prophylaxis	
MTX + CSP	**86**
MMF + CSP	**10**
Other†	**5**
CMV status, patient/donor‡	
+/+	**20**
+/−	**27**
−/+	**12**
−/−	**40**

*In 34 patients, high-resolution typing was performed: 27 were HLA-identical and 7 were not; 9 donors were matched by low/intermediate-level typing (see text for details).
†One patient received MTX, 3 patients received MTX + CSP + FK506, and 1 patient received MTX + CSP + MMF; 3 syngeneic recipients received no prophylaxis.
‡Information was missing in 5 patients.
§ATG/thymoglobulin.

Hematopoietic Cell Transplantation

Transplantation characteristics are summarized in Table 2. The conditioning regimens used were determined by sequential protocols that were active at the time of HCT. A total of 59 patients were conditioned with busulfan (BU) 16 × 1 mg/kg orally over 4 days, with dose adjustments made to achieve steady-state plasma levels of 800–900 ng/mL, plus CY 120 mg/kg (tBUCY). Six patients received BU 16 mg/kg (without targeting) plus CY 120 mg/kg (BUCY). Fifteen patients received total body irradiation (TBI) 6 × 200 cGy in combination with either BU 7 mg/kg (9 patients) or CY 120 mg/kg (6 patients), as described previously [5,11-14]. Fourteen patients (all in the new cohort) received tBUCY plus thymoglobulin (THY) 4.5 mg/kg over 3 days [15]. Nine patients (all in the new cohort) received nonmyeloablative conditioning with fludarabine (Flu) 90 mg/m^2 intravenously (IV) and TBI 200 cGy, because of older age (range, 41–70 years; median, 60 years) or concurrent nonhematologic disease [16], and 1 patient received Flu 120 mg/m^2 IV over 4 days, along with tBU [17].

A total of 43 patients received marrow, and 61 received G-CSF–mobilized peripheral blood progenitor cells (PBPCs) as a stem cell source. Graft-versus-host disease (GVHD) prophylaxis consisted of cyclosporine (CSP) and methotrexate (MTX) in 86 patients, mycophenolate mofetil (MMF) and CSP in 10 patients, MTX only in 1 patient, and various combinations of MTX or MMF and a calcineurin inhibitor in 4 patients [18,19]. Three syngeneic transplant recipients received no GVHD prophylaxis.

Criteria for Engraftment and Response

Time to engraftment was defined by the first of 3 consecutive days on which the absolute neutrophil count reached 0.5 × 10^9 /L [20]. Chimerism determinations on marrow cells were carried out at 28 and 100 days posttransplantation, again at 1 year posttransplantation, and at irregular intervals thereafter. GVHD was diag-

nosed based on previously established criteria [21,22]. Survival time was the time from transplantation until death or date of last contact (at which time patients were censored). Relapse-free survival was the time from transplantation until relapse or death from non–relapse-related causes. Relapse was defined as the reappearance of host cells and morphologic criteria of myelofibrosis after initial clearing of the marrow or the detection of previously existing cytogenetic abnormalities [5]. In some patients, relapse could not be determined definitively, because they showed reemergence of host cells (mixed chimerism) but no evidence of disease recurrence (see Results).

Determination of Comorbidities

Comorbid conditions were assessed by a comprehensive review of medical records in a computerized database. Scores were assigned by a single investigator (M.S.) blinded to outcome data, using the HCT-specific comorbidity index (HCT-CI) [16] developed by modifying the Charlson Comorbidity Index [23]. The modifications included a refinement of some comorbidity definitions by incorporating laboratory data, adding new comorbidities, and calculating new weights for comorbidities relevant for HCT recipients. The HCT-CI was shown to have greater sensitivity and higher predictive power for HCT outcomes than the original index [16].

Statistical Analysis

The association of various baseline characteristics with the hazard of overall mortality was examined by fitting Cox regression models [24]. Estimates of overall survival were obtained using the Kaplan and Meier method [25]. All 2-sided P values from regression models were estimated from the Wald test, with no adjustments made for multiple comparisons. Results were analyzed as of June 30, 2005.

RESULTS

Engraftment

A total of 101 patients could be evaluated for engraftment. Among allogeneic HCT recipients conditioned with regimens other than Flu + TBI (nonmyeloablative), the median time to granulocyte engraftment was 19 days. Patients who received PBPCs engrafted an average of 5.7 days earlier than those who received marrow ($P = .002$). Patients who underwent splenectomy engrafted an average of 2.6 days earlier than those with intact spleens ($P = .18$); and this difference was increased to 3.6 days ($P = .05$) after adjusting for source of stem cells. The difference between PBPCs and marrow did not change significantly after adjusting for splenectomy.

Graft Failure and Relapse

Seven patients experienced graft failure as documented by declining blood cell counts and reemergence of host cells in blood and marrow after initial engraftment. Four received transplants from HLA-nonidentical donors (3 family members and 1 unrelated). Three of these patients were conditioned with tBUCY and 1 was conditioned with CYTBI, and all 4 received marrow as a source of stem cells. The remaining 3 patients received transplants from HLA-identical unrelated donors, 2 after nonmyeloablative conditioning and 1 after tBUCY plus thymoglobulin; all 3 received PBPCs. Three among the 7 patients with graft failure experienced relapse, 3 died of non–relapse-related causes, and 1 was alive with autologous marrow recovery and no evidence of disease.

Another 6 patients were mixed chimeras at the 3-month evaluation. Among these, 2 subsequently lost their grafts and relapsed, and 2 converted to full donor chimerism and were alive in remission. Two patients remained mixed chimeras, with 100% $CD33^+$ cells at 2 years after HCT and 7%–34% $CD3^+$ donor cells at 2 and 11 years after HCT, respectively.

Thus, a total of 11 patients did not achieve sustained complete donor cell engraftment. Among these, 3 were alive with normal hematopoietic parameters (2 mixed chimeras and 1 with autologous recovery), 5 experienced relapse, and 3 died from non–relapse-related causes. In addition, 4 patients relapsed after having achieved complete donor cell chimerism. Of the 11 patients with relapse overall (including the 2 patients surviving with mixed chimerism), 9 had CIMF, 1 had PV, and 1 had ET. One of the 11 patients who relapsed achieved complete remission while developing GVHD on tapering CSP and survived in remission, whereas 8 patients died with relapsed disease.

GVHD

Overall, 65 of 99 evaluable patients (64%) given allogeneic transplants developed acute GVHD (grade II in 44, grade III in 19, and grade IV in 2), for incidences of 58% (29 of 50) among recipients of HLA-identical sibling transplants and 73% (36 of 49) among unrelated (both HLA-identical and nonidentical) or mismatched family member transplants. There was a trend toward less GVHD in HLA-identical sibling transplants ($P = .11$). Chronic GVHD occurred in 85 patients (84%) and was extensive, requiring therapy, in 62 (59%). No statistically significant difference was seen between HLA-identical sibling (56%) and unrelated donor or mismatched family member transplants (59%).

Table 3. *Causes of Death*

Causes of Death	Number of Patients
Progressive disease/relapse*	**8**
Nonrelapse causes	**33**
Pneumonia/pulmonary failure	**12**
MOF/HUS/TTP	**5**
Invasive aspergillosis ± GVHD	**7**
GVHD†	**6**
Miscellaneous‡	**3**

MOF indicates multiorgan failure; HUS, hemolytic uremic syndrome; TTP, thrombotic thrombocytopenic purpura.
*Two patients relapsed with lymphoma (see text).
†Including 1 patient with bronchiolitis obliterans.
‡Two patients died of sepsis, and 1 patient died of viral encephalitis.

Causes of Death

Causes of death are summarized in Table 3. Eight patients (1 syngeneic transplant and 7 allogeneic transplant recipients) died from relapse, and 33 died from non–relapse-related causes, 14 of these within 100 days of transplantation (day 100 mortality, 13%). The projected non–relapse-related mortality at 5 years was 34% (95% confidence interval [CI] = 28%–48%). The median time to relapse was 1.8 years.

Survival and Disease Response

A total of 63 patients were alive at last contact, for an estimated probability of survival of 61% (95% CI = 43%–65%) at 5 years (Figure 1A). Among these, 57 were conditioned with myeloablative regimens and received transplants from allogeneic donors, 1 received a syngeneic transplant, and 5 underwent transplantation after nonmyeloablative conditioning. Univariate regression models for overall mortality are summarized in Table 4. Patients conditioned with tBUCY had a higher probability of survival (68%) than those conditioned with other regimens (46%; hazard ratio [HR] for mortality = 0.4; 95% CI = 0.2–0.7; P = .001) (Figure 1B). Patients who had received antithymocyte globulin (ATG) in addition to tBUCY (n = 14) had a higher HR for mortality than those conditioned with tBUCY without added ATG (HR = 1.92; 95% CI = 0.74–4.98), although the difference was not statistically significant (P = .18). Of the 13 patients receiving transplants from HLA-nonidentical (related or unrelated) donors, 7 survived for a probability of 51% at 3 years. Among 9 patients conditioned with a low-intensity regimen of fludarabine and TBI, 5 (56%) are surviving. Patients in higher Dupriez risk groups had higher mortality rates, and patients with less extensive fibrosis tended to fare somewhat better than patients with more extensive fibrosis, although the difference was not statistically significant (Table 4). Patients with leukemic transformation tended to have a higher mortality rate than those without, although the numbers were small, and the difference did not achieve statistical significance (HR = 2.1; 95% CI = 0.8–5.3; P = .12). Results in 92 patients without leukemic transformation who received allogeneic transplants after myeloablative regimens are summarized in Figure 2. Mortality was slightly higher in patients receiving transplants from unrelated donors (HLA-identical by high-resolution typing) than in patients receiving transplants from genotypically HLA-identical siblings, but the difference was not statistically significant (HR = 1.4; 95% CI = 0.6–3.4; P = .42; Figure 3). After adjusting for patient age, HCT-CI, and conditioning regimen, the magnitude of the difference was reduced to HR = 1.2.

Patients with an underlying diagnosis of PV or ET had a significantly lower hazard of mortality than patients with other diagnoses (P = .03; Table 4) and hence superior survival, as shown in Figure 4. In a multivariable analysis, conditioning with tBUCY continued to be associated with a decreased hazard of mortality (Table 5). In addition, because there was a correlation between the underlying diagnosis and platelet counts (only 3 of 34 patients with low platelet

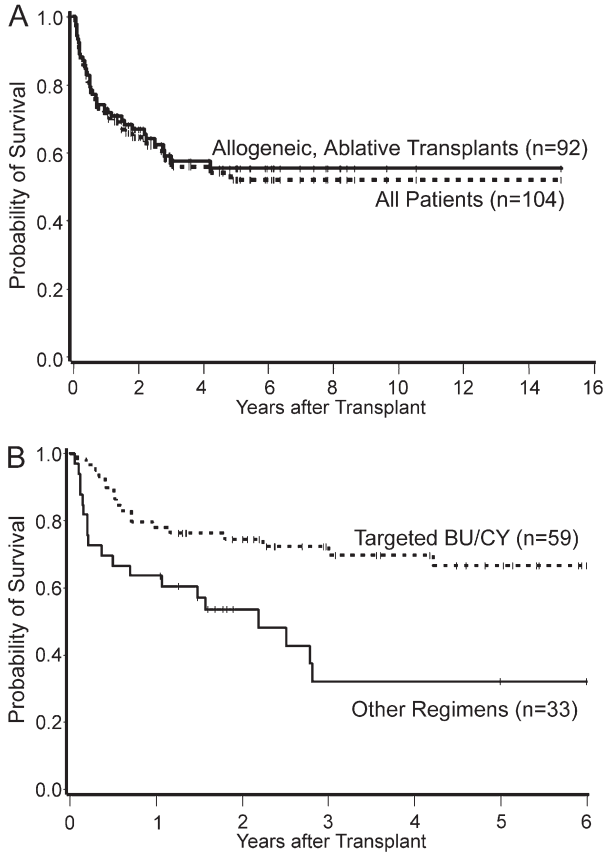

Figure 1. Overall survival. (A) All patients in the study (n = 104) compared to patients given allogeneic transplants after myeloablative conditioning regimens (n = 92). (B) Survival among the 92 allogeneic recipients prepared with myeloablative regimens by type of regimen.

Table 4. *Univariate Regression Models for Overall Mortality*

Factor (Number of Patients Dying/All Patients)	Hazard Ratio‡	95% CI	P
Conditioning regimen*			
Other regimens (24/45)	1	—	—
Targeted BUCY (18/59)	0.4	0.2–0.7	.001
Conditioning regimen†			
Targeted BUCY (18/59)	1	—	—
Other ablative regimens (20/36)	2.6	1.4–4.9	.003
Nonablative (4/9)	2.9	1.1–7.8	.04
Platelet count			
100×10^9/L or higher (25/70)	1	—	—
$< 100 \times 10^9$/L (17/34)	2.1	1.1–3.8	.02
Dupriez risk classification			
Low (11/44)	1	—	—
Intermediate (19/40)	2.3	1.1–4.9	.02
High (12/20)	2.8	1.2–6.3	.01
Dupriez risk classification			
Low	1	—	—
Intermediate/high	2.5	1.3–4.9	.009
Age, modeled as a continuous linear variable	Increasing age, increasing hazard	—	.01
Age, by cohort			
Younger than 40 (6/22)	1	—	—
40–50 (14/35)	1.5	0.6–3.8	.35
Older than 40 (22/47)	2.1	0.9–5.0	.09
Degree of fibrosis			
1–2 (11/34)	1	—	—
3 (14/28)	1.9	0.9–4.2	.11
4 (15/37)	1.5	0.7–3.2	.33
Karyotype			
Clonal (21/45)	1	—	—
Normal (20/53)	0.8	0.37–1.26	.22
Splenectomy			
No (32/69)	1	—	—
Yes (9/29)	0.6	0.3–1.2	.16
Source of stem cells			
PBPC (23/61)	1	—	—
Marrow (19/43)	1.1	0.6–2.0	.76
Donor			
Related HLA-identical or syngeneic (20/55)	1	—	—
Unrelated or mismatched related (22/49)	1.4	0.8–2.5	.28
Disease duration, modeled as continuous linear variable	—	—	.53
Evidence of transformation to leukemia			
No	1	—	—
Yes	2.1	0.8–5.3	.12
Diagnosis			
PV/ET	1	—	—
Other diagnoses	2.4	1.1–5.5	.03
HCT-CI, modeled as continuous linear variable	Increasing score, increasing hazard	—	.02
HCT-CI			
0–3	1	—	—
4–6	2.6	1.3–5.6	.01

*tBUCY versus all other regimens.
†Separate consideration of patients conditioned with a nonmyeloablative regimen.
‡HR < 1 indicates superior outcome; >1, inferior outcome.

counts carried a diagnosis of PV or ET), we considered the following variables: PV or ET plus high platelet count, diagnoses other than PV or ET plus high platelet count, and any diagnosis with low platelet count (Table 5). In this comparison among patients with high platelet counts, patients with PV or ET had a lower hazard of mortality than patients with other diagnoses, and both had lower hazards compared with patients with low platelet counts (HR = 0.3; 95% CI = 0.1–0.8; P = .01 and HR = 0.7 95% CI = 0.4–1.5; P = .39; Table 5). The small number of patients with an HCT-CI of 4–6 (n = 12) had a higher risk of death than did those with a score of 0–3 (HR = 2.3; P = .03; Table 5). After adjusting for conditioning regimen, patient age, HCT-CI, and the diagnosis/platelet count variable, the impact of Du-

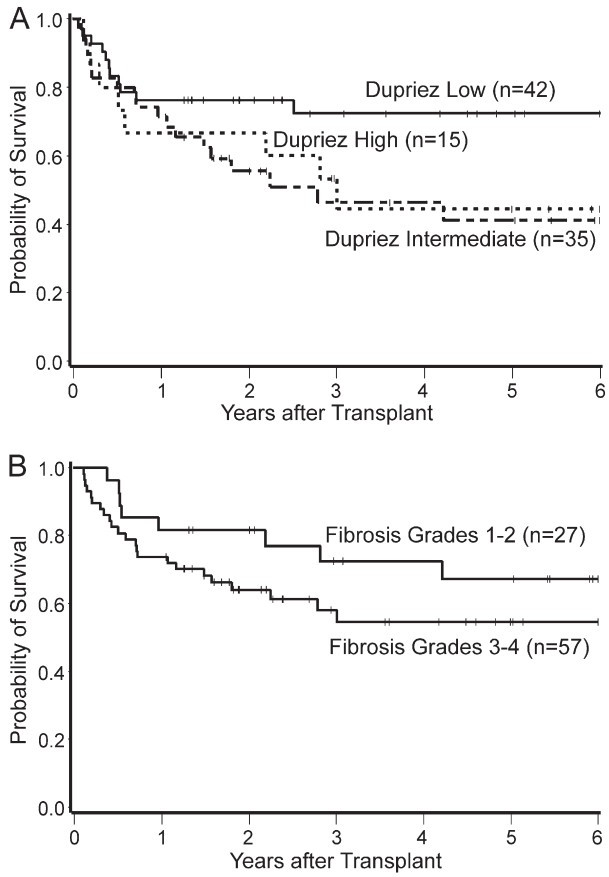

Figure 2. Impact of disease parameters on survival (same patients as in Figure 1). (A) Effect of Dupriez risk category. (B) Effect of the degree of marrow fibrosis (not graded in 5 patients).

priez risk group was no longer statistically significant ($P = .28$; modeled as continuous variable).

Hematologic parameters in surviving patients (last available determination at 5 months–11 years after HCT) included white blood cell counts of 1.9–25 $\times$ 10^9/L (median, 6.2 $\times$ 10^9/L), absolute neutrophil counts of 0.3–9.0 $\times$ 10^9/L (median, 3.3 $\times$ 10^9/L), platelet counts of 50–1167 $\times$ 10^9/L (median, 190 $\times$ 10^9/L), and hemoglobin levels of 9–17 g/dL (median, 12.6 g/dL). The lowest blood cell counts were seen in patients with the shortest follow-up and those with mixed chimerism.

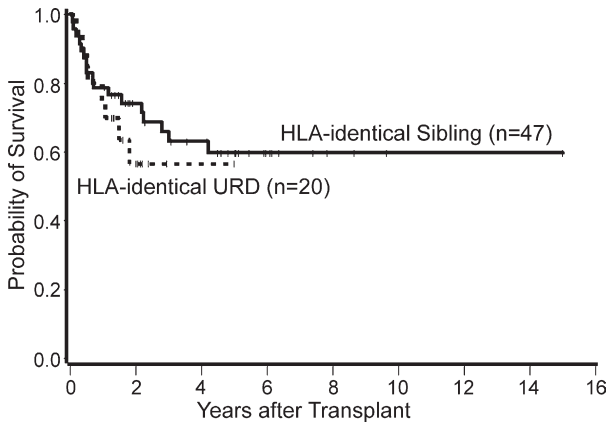

Figure 3. Overall survival among patients without leukemic transformation conditioned with myeloablative regimens and transplanted from HLA-identical sibling donors (n = 47) or unrelated donors who were HLA-identical by high-resolution typing (n = 20).

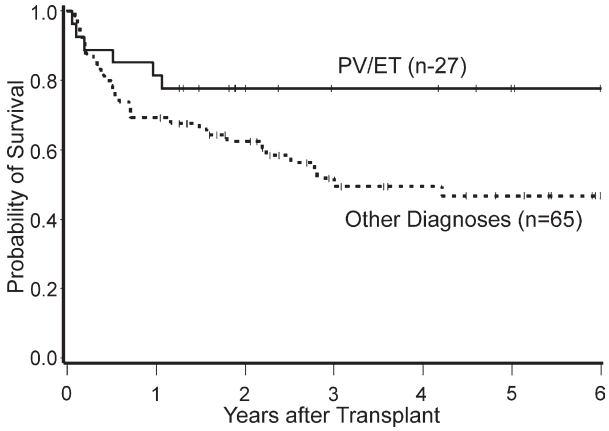

Figure 4. Impact of primary diagnosis on transplantation outcome (same patients as in Figure 1).

Posttransplantation marrow biopsy specimens were available in 82 patients surviving beyond 100 days. Among these, 42 showed no fibrosis or only traces of fibrosis; 19 were staged as grade 1 fibrosis, 14 as grade 2 fibrosis, and 5 as grade 3–4 fibrosis. In 2 patients, residual osteosclerosis was present but with no evidence of reticulin fibrosis. In the presence of 100% donor cell chimerism and normal blood cell counts or transfusion independence, the persistence of fibrosis (or osteosclerosis) by itself was not considered to represent relapse or persistent disease.

Among 66 patients with intact spleens and splenomegaly who survived beyond 100 days after transplantation, the spleen was of normal size in 50 patients at the time of the latest examination, between 6 months and 3 years after HCT. In 4 patients, the spleen was palpable at the costal margin; in 2 it was enlarged but < 8 cm, and in 4 it was > 8 cm. In 3 patients, the spleen was reported to be palpable, but no size was recorded, and in 3 other patients, no information on spleen size was available.

DISCUSSION

The present study confirms the curative potential of HCT for patients with myelofibrosis, with complete remission now extending up to 15 years in some patients. Survival was comparable in patients receiving transplants from HLA genotypically identical siblings and those receiving transplants from HLA-identical unrelated donors. The number of HLA-nonidentical transplants was too small to allow us to draw firm

Table 5. *Multivariable Regression Models for Overall Mortality*

Factor	Hazard Ratio	95% CI	P
Conditioning regimen			
Other regimens	**1**	**—**	**—**
Targeted BUCY	**0.5**	**0.3–0.9**	**.03**
Diagnosis/platelet count			
Low platelet count	**1**	**—**	**—**
PV/ET, high platelet count	**0.3**	**0.1–0.8**	**.01**
Other diagnoses, high platelet count	**0.7**	**0.4–1.5**	**.39**
Age, continuous linear variable	**Increasing age, increasing hazard**	**—**	**.06**
HCT-CI			
0–3	**1**	**—**	**—**
4–6	**2.3**	**1.1–4.9**	**.03**

Additional variables, such as Dupriez score, splenectomy, normal karyotype, degree of fibrosis, unrelated donor, stem cell source, and disease duration, did not statistically significantly improve the model.

conclusions, however. As in numerous other studies, the probability of successful transplantation declined with increasing patient age [26,27].

Similar to our previous report, Dupriez risk group was a significant factor for transplantation outcome in univariate analysis, although differences between the 3 risk groups were not statistically significant in a multivariable regression model. However, this observation does not exclude the possibility that Dupriez risk affected transplantation outcome, because the Dupriez parameters are likely to interact with other parameters, such as pre-HCT platelet count, which has been shown to be of prognostic significance by Dingli et al. [8] and to be correlated with survival after HCT in the present study. Therefore, clinical changes reflected in Dupriez risk group should be monitored, and progression should lead to the consideration of transplantation. Moreover, because 1/3 of patients in the present study carried diagnoses other than CIMF (for which the Dupriez classification was developed), different categorizations of disease groups may be indicated. In fact, a high pretransplantation platelet count, particularly in patients with PV or ET, was associated with a significantly reduced overall mortality. This finding was in agreement with observations in our initial report, where the hazard of transplantation-related mortality correlated inversely with platelet counts at the time of HCT [5].

Another disease parameter previously shown to affect posttransplantation prognosis was the degree of marrow fibrosis [4,5]. The present analysis again suggested such an effect, although it was less prominent than described initially (possibly related to the fact that there were 4 deaths from various causes among the 9 patients with the lowest degree of marrow fibrosis) [4,5]. Nevertheless, the role of fibrosis in transplantation outcome remains controversial in view of the considerable variation in sampling by single marrow biopsies and various classification schemes used [10,28]. Methods that assess larger volumes of marrow, such as magnetic resonance imaging (MRI), may improve the ability to more reliably classify marrow fibrosis [29]. Indeed, a recent report suggests that MRI is useful not only in assessing the extent of fibrosis, but also in monitoring regression of fibrosis after HCT [30].

As expected, patients who underwent transplantation because of leukemic transformation did less well than patients who did so because of peripheral blood cytopenias associated with marrow fibrosis. However, the major causes of death were organ toxicity and infections, rather than relapse, conceivably because these patients had received pretransplantation cytotoxic therapy. Alternatively, these patients had more advanced disease with associated extramedullary hematopoiesis, which is known to be associated with reactive fibrosis in the liver and lung, rendering those organs susceptible to regimen-related toxicity.

In agreement with previous data, there was an advantage (insignificant) for the tempo of engraftment in splenectomized patients, an effect that was more prominent after adjusting for the source of stem cells. In patients with intact spleens, there was no significant correlation between spleen size and the tempo of engraftment. Neither overall survival nor progression-free survival was affected by splenectomy. As in our initial report [5], the present data do not support the routine use of preemptive splenectomy as a strategy to improve HCT outcome.

The prognostic relevance of clonal cytogenetic abnormalities in patients with CIMF has been a matter of debate. A recent report by Tefferi et al. [31] suggests that the presence of del (20q) or del (13q) in CIMF carries a favorable prognosis. In the present study, clonal cytogenetic abnormalities did not significantly affect overall outcome. Of 8 patients with del (20q) or del (13q), 6 are surviving in remission; these numbers are too few to allow a firm conclusion. The recently described activating mutation (V617F) in JAK2, which is located on chromosome 9p [32], and for which data were not available in the present analysis, may affect the tempo of progression of fibrosis

and may have a more profound effect on prognosis than clonal chromosomal findings that have been investigated in the past [32,33].

Finally, older patient age was associated with significantly increased posttransplantation mortality. Because patients with myelofibrosis tend to be in an age range in which high-dose therapy generally is not well tolerated, this is an important observation in regard to the selection of transplantation conditioning regimens. As shown in earlier trials [11,34], the conditioning regimen had a significant effect on HCT outcome, with the highest success rate obtained with a targeted BUCY regimen. Overall survival with this regimen in the new cohort of patients was not statistically significantly different from that in the originally reported cohort ($P = .28$), even though the median age in the new cohort was almost 1 decade older. Nevertheless, non–relapse-related mortality was 13% at day 100 and 34% at 5 years, raising the question as to whether results could be further improved with the use of lower-intensity/nonmyeloablative conditioning regimens. Rondelli et al. [7] recently reported 80% survival in 22 patients with myelofibrosis who underwent transplantation at several institutions after conditioning with Flu, Cy, TBI, melphalan, or thiotepa in various combinations. Kroger et al. [6] presented data from a pilot study in 21 patients with myelofibrosis ranging in age from 32 to 63 years (median, 53 years) conditioned with BU 10 mg/kg plus Flu 180 mg/m^2 and ATG and receiving transplants from related or unrelated donors. With a follow-up of 4–59 months, the estimated 3-year relapse-free survival was 84%. These are very encouraging data. A nonmyeloablative regimen of Flu/TBI, as used in the present study, yielded a somewhat lower survival rate (56%), which did not differ from that achieved with more conventional regimens. However, this small cohort comprised patients up to age 70, most of whom had comorbid conditions [16] not present in the patients reported by Kroger et al. [6]. That comorbid conditions are relevant to the success of transplantation is supported by the current analysis, which showed a significant effect of the HCT-CI on survival; even among the 9 patients conditioned with a nonmyeloablative regimen, 3 died from transplantation-related complications. It is also important to note that the actual intensities of the conditioning regimens used in these studies differed. Conceivably, conditioning regimens of intermediate intensity will offer optimum results [35]. However, retrospective comparisons have yielded equivocal results [36-38], and only prospective trials will be able to determine the optimum approach [39].

Overall, 33 patients in the present study died from non–relapse-related causes for a cumulative incidence of 34% at 5 years, similar to previous reports [4,5]. Disease recurrence, on the other hand, was a relatively infrequent cause of death (7%). The estimated probability of relapse was 11% at 5 years, an indication that the conditioning regimens used along with the allogeneic effects of donor cells were effective in eradicating the disease in most patients. However, a few patients became long-term mixed chimeras. Cytogenetic markers were not available in these patients, and it was not possible to determine whether the original disease clone was present. It is not clear whether or not the persistent host cells have malignant potential, and further observation is needed.

Thus, this study, with follow-up extending to 15 years after HCT, confirmed that HCT offers potentially curative therapy for patients with myelofibrosis of various etiologies. The optimum timing for transplantation remains to be determined, although the data suggest that higher success rates can be expected at earlier disease stages. Indicators such as Dupriez classification may help in determining the optimum timing of transplantation. However, although earlier HCT tends to result in better outcome, patients with lower Dupriez scores also may enjoy longer survival with conservative management [2,40]. Monitoring of patients with CIMF for disease progression by, for example, sequential determination of peripheral blood CD34$^+$ cell counts [41], which were not available in the present analysis, or MRI of the marrow for changes in the extent of fibrosis, may reveal disease progression and facilitate the decision in terms of HCT. The optimum conditioning regimen may depend on patient age and comorbid conditions [42]. To what extent recent insights into the role of activating mutations in the tyrosine kinase JAK2 in patients with myeloproliferative disorders will affect treatment decisions remains to be determined.

In summary, although questions remain in terms of the timing of HCT and optimization of conditioning regimens, HCT clearly has curative potential for patients with myelofibrosis and promotes long-term survival in remission.

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