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Severe Infections after Unrelated Donor Allogeneic Hematopoietic Stem Cell Transplantation in Adults: Comparison of Cord Blood Transplantation with Peripheral Blood and Bone Marrow Transplantation

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ABSTRACT

We evaluated the occurrence of severe infections in 192 consecutive adult recipients of volunteer unrelated donor allogeneic hematopoietic stem cell transplants, with a detailed analysis of severe infections after receipt of cord blood transplants (CBTs; n = 48) or bone marrow transplants (BMTs)/peripheral blood stem cell transplants (PBSCTs; n = 144). At a 3-year median follow-up, CBT recipients had a higher risk of developing any severe infection (85% versus 69% in BMT/PBSCT recipients, $P < .01$). CBT recipients had a higher incidence of severe bacterial infections before day +100, but at 3 years the risks of these and other infections were similar in the CBT and BMT/PBSCT groups. In addition, the 100-day and 3-year incidences of infection-related mortality (IRM) did not differ between groups ($P = .2$ and .5, respectively). In multivariate analysis, the most significant risk factor for IRM in all 192 patients was monocytopenia ($.2 \times 10^9$/L). In CBT recipients, only neutropenia ($.2 \times 10^9$/L) on day +30 and low nucleated cell dose infusion ($<2 \times 10^7$/kg) showed a trend for increased IRM ($P = .05$ in both cases). Stem cell source had no effect on day +100 or 3-year nonrelapse mortality (NRM), cytomegalovirus infection, cytomegalovirus disease (7% versus 6%), or overall survival (36% versus 39%, respectively). The number of mismatches in HLA (A, B, and DRB1) had no effect on any outcome in CBT recipients. In contrast, in the BMT/PBSCT group, the presence of any mismatch by low or high-resolution HLA typing (A, B, C, and DRB1) increased NRM and decreased overall survival ($P < .01$). IRM was the primary or secondary cause of death in 61% and 59% of CBT and BMT/PBSCT recipients who died, respectively. Our results confirm the relevance of severe infectious complications as source of severe morbidity and NRM after volunteer unrelated donor hematopoietic stem cell transplantation in adults, but suggest that CBT recipients have a similar risk of dying from an infection if an accurate selection of a cord blood unit is done.

KEY WORDS

Severe infections • Cord blood • Unrelated hematopoietic stem cell transplantation

INTRODUCTION

Serious infections are common problems after volunteer unrelated donor (VUD) hematopoietic stem cell transplantation (HSCT) and account for substantial morbidity and mortality, especially in adults [1-8]. Nonrelapse mortality (NRM) due to infections has an incidence as high as 45% to 60% only in the first year after HSCT [5,9,10,11]. Until recently, VUD HSCT used bone marrow transplants (BMTs) or peripheral blood stem cell transplants (PBSCTs). In recent years, however, umbilical cord blood transplantation (CBT) has been considered a valid alternative to BMT and PBSCT, and recent retrospective studies have suggested that CBT in adults has similar outcomes as BMT from VUD, provided a high stem cell dose is infused [12-14].

However, severe infections may be more common after CBT than after BMT/PBSCT, in large part attributable to the slower hematopoietic and immune reconstitutions after CBT [12,13,15]. A recent retrospective study in children has shown that the risk of severe infections after pediatric CBT is comparable to that of non–T-cell–depleted BMT and lower than that of T-cell–depleted BMT [16]. Similar studies are scarce in adults and have been reported with a short follow-up [11-14,17].

For this purpose we compared in detail the occurrence of severe infections after VUD transplantation with BMT or PBSCT with recipients of a CBT protocol in 192 adults, with special emphasis on infection-related mortality (IRM).

METHODS

Patient Selection

The present study was done by members of the Infectious and Non-infectious Subcommittee of the Spanish Group for Hematopoietic Transplantation (GETH). GETH member centers that perform >10 VUD allogeneic HSCTs per year were invited to participate in this study. To reflect the reality seen in clinical practice and have ≥1-year follow-up for all patients, centers had to complete an extensive case-report form for all consecutive patients who were >15 years old and received a VUD transplant with any source of stem cells between January 1997 and March 2004. Seven centers agreed to participate, and the final database was closed in June 2005.

In accord with local, European, and international legislation, all patients signed an informed consent before transplantation, which included authorization for the reporting and use of their data (assuring confidentiality) for current or future studies. Specific transplantation protocols also had local and national authorization with institutional review board approval.

Patient characteristics and noninfectious transplantation outcomes are presented in detail in Table 1. As expected, patients' characteristics differed in various baseline variables between stem cell transplantation groups, as described in recent registry studies [12-14].

Transplantation Procedure and Supportive Care

Before hematopoietic engraftment, all patients were isolated in single rooms with high efficiency particulate air filtration (n = 150) or laminar airflow (n = 42). All patients received systemic antibiotic prophylaxis, starting before transplantation, which consisted of a fluoroquinolone in 86% of cases. In addition, during neutropenia antiviral and antifungal prophylaxes consisted of acyclovir and fluconazole in >90% of patients.

High-dose acyclovir from days −4 to +30 was used as cytomegalovirus (CMV) prophylaxis immediately after HSCT in 46 patients (24%). Seven patients (3.5%) continued prophylaxis with ganciclovir or foscarnet even after engraftment and until 100 day after HSCT. In all other patients, preemptive therapy with the latter 2 drugs was used. Both strategies were used in similar proportions in CBT and BMT/PBSCT recipients (pre-emptive therapy was used in 70% and 81% transplant groups, respectively). Monitoring of CMV infection was done at least weekly with detection of pp65 antigenemia in peripheral blood after engraftment in all cases, and qualitative or quantitative polymerase chain reaction for CMV DNA was also used in 20% of patients (all after 2001). In a similar manner, serum galactomannan was used routinely only since 2002, so results of polymerase chain reaction–based diagnosis of CMV infection and galactomannan antigenemia for the diagnosis of invasive aspergillosis (IA) were not analyzed in this study because their inclusion could lead to a bias showing a falsely higher number of CMV infections and probable IA in the latter years.

After engraftment, 125 patients (73% of evaluable patients on day +30) received cotrimoxazole for prophylaxis against *Pneumocystis jiroveci* and toxoplasmosis. One hundred eleven patients (58%) received intravenous immunoglobulin until day +100. There were no differences in supportive care procedures between transplantation groups (data not shown).

Definitions

As defined in most studies, severe infections included those potentially associated with death or severe clinical compromise, which is described in detail below. Other infections not requiring therapy or those requiring only oral antibiotics on an outpatient basis were excluded from this study.

A pre-established homogeneous set of definitions was used by all participating centers, which followed the proposed guidelines from the Infec-

Table 1. *Patient Characteristics and Main Transplantation Outcomes (n = 192)*

Transplant-Related Characteristics	CBT (n = 48)	BMT (n = 120)	PBSCT (n = 24)	P
Median age, range	31 (16-47)	30 (16-59)	37 (16-54)	.06
≥35, y	18 (38)	38 (32)	14 (58)	.05
Male	30 (62)	64 (54)	16 (67)	
Sex mismatched	22 (52)	48 (40)	11 (46)	
Female donor/male recipient	13 (27)	20 (17)	7 (29)	
Underlying disease				
AL/MDS	29 (60)	60 (50)	16 (67)	
CML	19 (40)	46 (38)	7 (29)	
Lymphoid malignancies	—	14 (12)	1 (4)	
Status at transplant*				
Early	24 (50)	72 (60.5)	12 (50)	
Not early	24 (50)	48 (40)	12 (50)	
Second transplant	9 (19)	28 (23)	5 (20.9)	
Status of CMV serology				
Recipient positive (high risk)	36 (75)	87 (73)	22 (92)	
Recipient negative (intermediate and low risk)	12 (25)	32 (27)	2 (8)	
Period of transplantation				
1997-2000	23 (48)	68 (57)	7 (29)	
2001-2004	25 (52)	52 (43)	17 (71)	
Level HLA testing available				
Class I HLA (A, B, C) by PCR-HR	1	78 (65)	19 (79)	<.001 for BM/PBSCT vs CBT
Class I HLA (A, B) by LR	45 (94)	42 (35)	5	
Class II HLA (DRB1 ± DQB1) by PCR-HR	48 (100)	120 (100)	24 (100)	
Number of known mismatches, if considering A and B by LR and DRB1 by HR				
6/6 match	5 (11)	110 (92)	20 (83)	<.001 for BM/PBSCT vs CBT, when comparing 6/6 match with any mismatch
7/8 match	NA	18/74 (24)	5/19 (26)	
8/8 match	NA	56/74 (76)	14/19 (74)	
5/6 match	25 (52)	7 (6)	3 (13)	
>1 mismatch	18 (38)	3 (3)	1 (4)	
High stem cell doses (see Definitions for details)	25 (52)	53/110 (48)	11/21 (52)	
Conditioning regimen				
Myeloablative conditioning	48 (100)‡	101 (84)	19 (79)	
TBI-based ablation		77 (64)	17 (71)	
Reduced intensity conditioning†	—	19 (16)	5 (21)	
ATG used as part of in conditioning	48 (100)	24 (20)	6 (25)	<.001 for BM/PBSCT vs CBT
GVHD prophylaxis				
CsA (± steroids) plus ATG	48 (100)	3 (3)	4 (17)	<.001 for BM/PBSCT vs CBT
CsA plus MTX plus ATG		21 (18)	2 (8)	
CsA plus MTX/MMF, without ATG		96 (80)	18 (75)	
Follow-up after transplantation, alive/died	17/31	45/75	12/12	
Time (mo) to death, median, (range)	2.4 (0.6-23)	3.3 (0.3-54)	1.7 (.6-6.2)	.05 for BMT vs CBT and PBSCT
Follow-up (mo) in survivors, median (range)	48 (31-95)	53 (21-95)	38 (16-62)	.05 for BMT and CBT vs PBSCT
3-Year NRM, % cumulative incidence (95% CI)	48 (23-59)	42 (33-51)	38 (12-51)	.3
3-year OS, % probability (95% CI)	36 (26-44)	40 (31-49)	38 (24-52)	.7
GVHD and hematopoietic recovery				
GVHD, n (% cumulative incidence)				
Grades 2-4/3-4 aGVHD	35 (45)/14 (18)	68 (41)/27 (22)	12 (40)/8 (29)	>.1/.1
Extensive cGVHD, n (% cumulative incidence)	6 (9)	23 (16)	4 (15)	.1
Hematologic recovery, × 10^9/L, median (range)				
Day to stable neutrophil engraftment (>0.5)#	22 (13-52)	19 (10-31)	16 (11-23)	Detailed below§
Patients with <.5 neutrophils at day +30	4 (8)	1 (1)	0	
Day to platelets >20#	67 (10-230)	21 (5-138)	12 (9-38)	
Day to monocytes >0.2	27 (13-66)	19 (10-366)	17 (10-23)	
Day to lymphocytes >0.2	41 (12-69)	23 (4-399)	17 (7-41)	
Day to lymphocytes >0.5	49 (26-147)	36 (8-269)	24 (8-51)	

Data are numbers (%) unless otherwise noted. AL indicates acute leukemia; MDS, myelodisplastic syndrome; CML, chronic myeloid leukemia; PCR-HR, DNA high-resolution HLA testing, to the allele level, by polymerase chain reaction; LR, serologic testing for A, B, or low-resolution PCR testing; TBI, total body irradiation; CsA, cyclosporine A; MTX, methotrexate; MMF, mycophenolate mofetil; ATG, antitymocyte globulin; GVHD, graft-versus-host disease; NRM, nonrelated relapse mortality; NA, not applicable.

*Early: Acute leukemia and myelodisplastic syndrome in first complete remission after chemotherapy (<5% blasts), previously untreated low-risk MDS, chronic myeloid leukemia in first chronic phase. Not early: Other disease status of previously mentioned hemopathies; multyple myeloma, chronic lymphoid leukemia, non-Hodgkin lymphoma, hodgkin disease, solid tumors.

†Included fludarabine-busulfan or fludarabine-melphalan, as previously described.

‡A homogeneous conditioning regimen was used for all cord blood transplants, with thiotepa, cyclophosphamide, busulfan and ATG, as previously described [33].

§For CBT vs BMT and PBSCT, all cell types recovered faster ($P < .01$) in the latter groups. For BMT vs PBSCT, all cell types recovered earlier in the latter ($P < .01$).

#Proportions of patients who did not achieve stable neutrophil engraftment/stable platelet engraftment by stem cell source were 3 (6%)/9 (19%) for CBT recipients and 1 (0.8%)/9 (7%) for BMT-PBSCT recipients, respectively.

tious Diseases Working Party of the European Group for Blood and Marrow Transplantation (http://www.ebmt.org/5WorkingParties/IDWP/wparties-id6.htm). These definitions included: (1) documented bacterial infection of any type and organ site, except bacteremia by coagulase-negative staphylococci, *Micrococcus* spp, and saprophytic *Corynebacterium* spp, which were not included in the present analysis; (2) invasive fungal infections (IFIs), which were divided into proved and probable according to currently accepted criteria [18], although galactomannan-defined probable IA was not included; (3) CMV infection, defined as the presence of a single positive antigenemia (> 1 pp65 antigen-positive leukocyte among 200 000 cells counted per slide), CMV viremia on the basis of a positive blood culture or shell vial centrifugation culture, or documentation of CMV disease without previous positive antigenemia or viremia [19], and CMV disease was defined as the demonstration of CMV in biopsy or autopsy specimens from clinically involved visceral sites by culture and/or histology or if CMV was detected in samples from clinically defined sites of disease [19]; (4) disseminated varicella-zoster virus infection, defined as cutaneous involvement of ≥2 skin dermatomes or visceral involvement; (5) other severe viral infections such as human herpesvirus 6 encephalitis, Epstein-Barr virus post-transplant lymphoproliferative disease, adenovirus disease, and lower respiratory tract infection by respiratory viruses were diagnosed according to criteria of the Infectious Diseases Working Party of the European Group for Blood and Marrow Transplantation [20-22] (detailed in Table 2); and (6) pneumonia of unknown origin, defined as any new radiologic lung infiltrate in a febrile patient with respiratory symptoms in the absence of a known pathogen.

Patients were considered to have died from infection (ie, IRM) if death was attributed to a recent severe infection by the primary investigator in each center and/or an infection was identified at autopsy. Patients with relapse of their underlying disease before death were excluded from this definition, even if an infection was the final cause of death, whereas those who died with clear evidence of a life-threatening infection were considered to have an IRM. All cases of IRM as considered by each participating center were verified by 1 of the study coordinators (R. Martino). However, a confirmatory autopsy would have been the only way of establishing the definite cause of death. As presented in Table 3, similar proportions of autopsies were done in patients with IRM in all stem cell source transplantation groups (21% to 25% of NRMs).

A significant number of patients with graft-versus-host disease (GVHD) under treatment developed a clinically defined infection before death (pneumonia in 85% of cases) with no microbiologic findings and often progressing to multiorgan failure despite the use of multiple antimicrobial agents. Although a probable or proved infection was not found and an autopsy was not granted by the patients' families, these deaths were considered as IRM of unknown pathogen(s).

The period after transplantation that any event occurred was classified as early (from days 0 to +30), intermediate (days +31 to +100), late (days +101 to +365, ie, +1 year), or very late (day +365 after transplantation).

Disease phase of the underlying disease at transplantation was categorized as early or not early, as defined in Table 1 (see notes). Assessment, grading, and treatment of acute GVHD (aGVHD) and chronic GVHD (cGVHD) were done using standard methods [23,24]. The stem cell dose infused was categorized separately in each transplantation group. The median stem cell dose per group was calculated, and stem cell doses above the median were categorized as high. A high level of stem cells was defined separately for each stem cell source: bone marrow ($>4 \times 10^8$/kg total nucleated cells (TNC) or $>4 \times 10^6$/kg $CD34^+$ cells), peripheral blood ($>6 \times 10^6$/kg $CD34^+$ cells), and cord blood ($>2 \times 10^7$/kg frozen TNC or $>1 \times 10^5$/kg frozen $CD34^+$ cells). The exact dose of cells given was not reported in 10 BMT recipients (8%) and 3 PBSCT recipients (12%).

Statistical Analysis

The major endpoint of this study was to compare the incidence of IRM in adults who received an unrelated donor HSCT with CBT versus BMT or PBSCT. Other outcomes analyzed in detail were NRM, CMV infection and disease, IFIs, pneumonia of unknown origin and overall survival (OS). The incidence of these outcomes (except OS) was analyzed with cumulative incidence, with the corresponding competing risk factors for each outcome, as described in detail elsewhere [25,26]. For OS the Kaplan-Meier estimate was used. Although showed in detail in Table 1, no attempt was made to analyze in detail risk factors for other important transplantation outcomes (relapse, disease-free survival, hematopoietic recovery, aGVHD, cGVHD, and others), because they were not the objects of this study.

All bacterial infections and other uncommon severe infections are presented in detail in the tables, and univariate analyses were done by pooling together all serious bacterial infections (but not multivariate models), due to the large number of different pathogens involved and the few cases per pathogen. Specifically, rates of serious bacterial infections before day +100 (early-intermediate periods) were analyzed by infection density (number of bacterial infections per 1000 patient-days).

To avoid erroneously finding statistically significant risk factors, only a prespecified set of variables was analyzed in univariate and multivariate testing,

Table 2. *Severe Infections in Each Transplantation Group by Period after Transplantation*

	Early (<Day +30)			Intermediate (Days +31 to +100)			Late (Days +101 to +365) and Very Late (after Day +365)			Total Severe Infections (All Stem Cell Sources, All Follow-up)		
	Total Cases	CBT	BMT/ PBSCT	Total Cases	CBT	BMT/ PBSCT	Total Cases	CBT	BMT/ PBSCT	Total Cases	CBT	BMT/ PBSCT
Patients at risk, n*	192	48	144	172	43	129	123	28	95	192	48	144
Patients infected in each period, n	62	26	36	79	17	62	63	21	42	140	41	99
Infectious episodes in each period, n	66			128			104			298		
Bacterial infections	32 (17)	15 (31)	17 (12)	23 (13)	3 (7)	19 (15)	26 (21)	9 (32)	17 (21)	72 (38)	31 (64)	77 (53)
Bacteremia†	28	12 (25)	16 (11)	27	2 (5)	25 (19)	21	7 (25)	14 (15)	76	21 (44)	55 (38)
Gram-negative bacteria	19	10 (21)	9 (6)	21			19			59		
Escherichia coli	8	6	2	6	—	6 (5)	5	1	4	19		
Other enterobacteria	2	1	1	6	—	6	7	2	5	21		
Pseudomonas aeruginosa	4	1	3	3	—	3	3	—	3	10		
Other GNFGNB	1	1	—	6	2	4	4	2	2	11		
Others GNB	4	1	3	—	—	—	—	—	—	4		
Gram-positive bacteria	9	2 (4)	7 (5)	6			2			17		
Staphylococcus aureus	4	1	3	1	—	1	1	1	—	6		
***Enterococcus* spp.**	1	—	1	2	—	2	1	1	—	4		
Viridans group streptococci	3	1	2	—	—	—	—	—	—	3		
Other GP bacteriae	1	—	1	3	—	3	—	—	—	4		
Pneumonia‡	5	4 (8)	1 (1)	10	1 (2)	9 (7)	8	2 (7)	6 (6)	23	7 (14)	16 (11)
Gram-negative bacteria	4			7			6			17		
Escherichia coli	—	—	—	—	—	—	1	—	1	1		
Other Enterobacteriaceae	—	—	—	2	—	2	1	—	1	3		
Pseudomonus aeruginosa	2	1	1	1	—	1	2	1	1	5		
Other GNFGNB*	2	2	—	4	1	3	2	—	2	8		
Gram-positive bacteria	1			3			2			6		
Staphylococcus aureus	1	1	—	2	—	2	1	1	—	4		
***Enterococcus* spp.**	—	—	—	—	—	—	—	—	—	—		
Viridans group streptococci	—	—	—	—	—	—	—	—	—	—		
Others GP bacteria	—	—	—	1	—	1	1	—	1	2		
Meningitis§	2	2	1	1	1	—	1	—	1	4	3	1
Other sites involved	1	—		—	—	—	4	—	4	5	—	5
Bacterial infection density‖		13	10		8	4						
Invasive fungal infections (n patients)	9 (5)	5 (10)	4 (3)	12 (7)	5 (12)	9 (7)	15 (11)	1 (4)	14 (15)	38 (19)	12 (23)	25 (17)
Candidemia¶	6	3	3	6	2	4	—	—	—	12 (6)	5 (10)	7 (5)
Invasive aspergillosis#	3	2	1	9	4	5	14	1	13	26 (14)	7 (15)	19 (13)
Invasive pulmonary aspergillosis	3	2	1	7	3	4		1	1			
Other sites	—	—	—	1	1	—		—	5 (1 v late)			
Other IFIs**							2		2	2		2
Toxoplasmosis disease (n patients)	—	—	—	4 (2)	2	2				4 (2)	4 (4)	—
Viral infections (n patients)	18 (9)	8 (17)	10 (7)	54 (31)	9 (21)	45 (35)	27 (22)	14 (50)	13 (14)	102 (53)	29 (60)	73 (51)
CMV												
Infection	11 (6)	7 (15)	4 (3)	52 (30)	9 (21)	43 (33)	12 (10)	4 (17)	8 (8)	75 (39)	20 (42)	55 (38)
Disease	1	—	1	11 (10)	3 (7)	8 (6)	10 (8)	5 (21)	5 (2 v late)	22 (11)	7 (17)	15 (10)

Table 2. *Continued*

	Early (<Day +30)			Intermediate (Days +31 to +100)			Late (Days +101 to +365) and Very Late (after Day +365)			Total Severe Infections (All Stem Cell Sources, All Follow-up)		
	Total Cases	CBT	BMT/ PBSCT	Total Cases	CBT	BMT/ PBSCT	Total Cases	CBT	BMT/ PBSCT	Total Cases	CBT	BMT/ PBSCT
HSV 1/2	3	1	2	2	1	1	7 (6)	1	6 (3 v late)	12 (6)	3 (6)	9 (6)
VZV (≥2 dermatomes)	1	—	1	1	—	1	22 (18)	11 (1 v late)	11 (5 v late)	24 (12)	11 (23)	13 (9)
HHV-6 encephalitis	1	—	1	1	—	1	1	1	—	3	1	2
Respiratory virus††	4	—	4	2	1	1	1	—	1	7	1	6
EBV lymphoproliferative disease	—	—	—	3	1	2	2	1	1	5	2	3

CB indicates cord blood; BM, bone marrow; PB, peripheral blood; GNFGNB, glucose nonfermenting negative gram bacteria; GNB, gram-negative bacillus; GP, gram-positive; CMV, cytomegalovirus; HSV, herpes simplex virus; VZV, varicella-zoster virus; HHV-6, human herpex virus 6; CRV, community respiratory virus; EBV, Epstein-Barr virus; v late, very late.

*Number of patients alive and without progression or relapse of their underlying malignancy at the start of each period analyzed. Groups of severe infection (bacterial, fungal, viral, and toxoplasmosis) are expressed as number of patients (percentage of patients at risk), but different subsets refer to infectious episodes that may not be coincident with number of patients, because there were patients that developed >1 severe infection.

†Bacteremias (number of episodes). Other Enterobacteriaceae include *Enterobacter cloacae* (4), *Proteus mirabilis* (3), *Serratia liquefaciens* (1), *Samonella* spp. (3), *Citrobacter freundi* (1). Other GNFNGB: *Acinetobacter* spp (3), *Stenotrophomonas maltophilia* (3), *Burkholderia cepacia* (1) and *Pseudomonas stutzeri* (1). Other GNB: *Fusobacterium nucleatum* (3), *Capnocytophaga* spp (1). Other GP bacteriae: *Leuconostoc* spp (1), *Clostridium difficile* (1), *Bacillus cereus* (1), *Listeria monocytogenes* (1).

‡Pneumonias (number of episodes). Other Enterobacteriaceae include: *Klebsiella pneumoniae* (1), *Salmonella* spp. (1), *Enterobacter cloacae* (1). Other GNFNGB: *Acinetobacter* spp (5), *Stenotrophomonas maltophilia* (5). Other GP bacteriae: *Streptococcus pneumoniae* (1), *Nocardia* spp (1).

§Meningitis: *Staphylococcus aureus* (1, early), *S. pneumoniae* (1, early), *Acinetobacter* spp. (1, intermediate), *Nocardia* spp (1, late).

‖Rates of serious bacterial infections before day +100 were analyzed by the infection density (number of bacterial infections per 1000 patient-days).

¶Includes *C krusei* (2), *C parapsilosis* (4), *C glabrata* (3), *C hemicoal* (1), *C tropicalis* (1), undetermined (2).

#Includes *A fumigatus (9)*, *A flavus (5)*, *A terreus (1)*, *A niger (3)*, *A candidus (1)*. Subgroup nonspecified in 9 cases.

**Mucormycosis, species not identified (1), *Penicillium marneffei* (1), *Pneumocystis carinii* (1).

††Respiratory syncytial virus (3), influenza virus A (1), adenovirus (3).

Table 3. *Overall Infectious Mortality and Causes of Death (Frequency with Respect to Total Patients Who Died in Parentheses, Unless Specified Otherwise)*

	CBT (n = 48)	BMT (n = 120)	PBSCT (n = 24)	P
Deaths, n	31 (65)	75 (62)	12 (50)	
100-Day IRM, % cumulative incidence (95% CI)	30 (10-40)	28 (19-37)	22 (9-25)	.2
3-Year IRM, % cumulative incidence (95% CI)	40 (12-58)	42 (33-51)	38 (12-51)	.5
Postmortem studies done, n (% NRM)	6 (21)	15 (25)	2 (22)	.8
IRM*	19 (61)	42 (56)	8 (67)	
Bacterial infection, n deaths/n cases (%)	9/31 (29)	9/69 (13)	4/8 (50)	
Pneumonia	4/7	8/14	2/2	
Meningitis	3/3	1/1	0	>.1 for all causes of IRM
Bacteremia ± other site(s)	2/21	0/54	2/6	
CMV disease	3/7	7/12	2/3	
Disseminated adenovirus disease	—	—	1/1	
Respiratory syncitial virus pneumonia	—	1/2	—	
Invasive fungal infections, n deaths/n cases (%)	7/11 (64)	19/23 (83)	1/2 (50)	
Invasive aspergillosis	5/7	16/17	1/2	
Disseminated candidiasis	2/5	3/7	0	
Toxoplasmosis	1/2	0	2/2	
Pneumonia of unknown etiology	5/7	18/28	4/6	
Severe GVHD + CDI ± MOF†	10 (32)	24 (32)	3 (25)	
Period after transplantation of IRM (%)				.2
Early (< day +30)	5 (26)§	3 (7)§	2 (25)§	
Intermediate (days +31 to +100)	9 (47)§	16 (38)§	4 (50)§	
Late (days +101 to +365)	4 (21)	19 (45)	2 (25)	
Very late (> day +365)	1 (5)	4 (9)	—	
Other causes of NRM	9 (29)	17 (23)	1 (4)	
Relapse of underlying malignancy‡	3 (6)	15 (12)	3 (12)	

BMT indicates bone marrow transplantation; PBSCT, peripheral blood stem cell transplantation; CBT, cord blood transplantation; IRM, infection-related mortality; NRM, nonrelapse mortality; GVHD, graft-versus-host disease; CDI, clinically documented infection; MOF, multiorgan failure.

*Total number of patients who died of IRM in each group. Different etiologies are not necessarily coincident with patients because death was linked to multiple causes in some of them.

†Included number of patients with a clinically defined infection before death associated with severe GVHD, leading to MOF despite the use of multiple antimicrobials. However, no pathogens were found at the time of autopsy, if performed.

‡Percentage refers to total number of patients in each group (alive and dead).

§In the 70 BMT/PBSCT recipients who were known to have an 8/8 allele match, IRM before day +100 was only 5% (4/70), much lower than in the 26 patients with a known mismatch (8/26, 30%) or CBT recipients ($P < .01$ for the comparison of fully matched BMT/PBSCT vs mismatched BMT/PBSCT or CBT).

when appropriate. All post-transplantation events or risk factors were entered in univariate and multivariate models as time-dependent covariates (Table 4). Cox proportional hazards regression was used to examine univariate and multivariate effects of each variable analyzed on the outcomes studied. Because the effect of the stem cell source group had a time-varying effect for the incidence of IFIs, especially IA, this outcome was divided into early and late events at an optimal time point as estimated by visual analysis of the cumulative incidence curves (around day +100).

Multivariate analyses included those variables with a P value <.1 in previous univariate testing. To analyze the effect of lymphocytopenia, monocytopenia, and neutropenia in each period, the counts reported on day +30 and those reported for patients still at risk at the beginning of each period were used. To compare characteristics of case patients with controls, summary statistics, including frequency counts and percentages for categorical variables, and medians and ranges were calculated. Comparisons from 2 × 2 tables were made by means of chi-square or Fisher exact t tests. The median times until onset of events were compared by means Wilcoxon rank-sum test. Cumulative incidence curves up to 3 years after transplantation were produced for all events analyzed, because the median follow-up for survivors exceeded 3 years in all study cohorts (detailed in Table 1). Quantitative variables were re-entered as qualitative or binary variables only if at least a trend for statistical significance ($P < .08$) was found. Tests of significance were 2-sided, with a significance level of $P \leq .05$. However, because the number of statistical tests being carried out was high for the sample sizes, final multivariate P values between .1 and .02 were considered as trends, whereas those <.01 were considered as truly statistically significant.

Before comparing CBT with the other stem cell sources, we analyzed the effect of BMT versus PBSCT on all outcomes. There were no differences in any outcome between these 2 latter groups, so these

Table 4. *Variables Tested in Univariate and Multivariate Models (if Applicable)*

Related to transplant
- **Graft type: bone marrow/peripheral blood stem cells vs cord blood transplantation**
- **Conditioning regimen: reduced intensity vs conventional myeloablation***
- **Year of transplant: 1997-2000 vs 2001-2004**
- **Hematopoietic stem cell dose (analyzed only for each stem cell source separately)**
- **Level of HLA matching:**
  - **A and B antigens by serology or low-resolution PCR and DRB1 hy high-resolution allele-specific PCR**
  - **A, B, DRB1, and DQB1 by high-resolution allele-specific PCR (analyzed only in bone marrow/peripheral blood stem cell transplant recipients)**
- **Use of antithymocyte globulin in the conditioning regimen**

Related to patient and disease
- **Patient age**
- **Gender and sex match: male patient to female donor vs other**
- **Second transplant, after failure of prior autologous hematopoietic stem cell transplantation: yes vs no†**
- **Disease status at transplantation advanced vs early†**
- **Pretransplant CMV risk group: patient positive/donor negative vs other combinations**

Post-transplantation variables‡
- **Use of steroids at ≥0.5 mg/kg for >2 wk: yes vs no§**
- **Use of ATG for treatment of GVHD§**
- **Severe neutropenia (<.2 × 10^9/L) at days +30, +90, +180, and +360**
- **Severe lymphocytopenia (<.2 × 10^9/L) at days +30, +90, +180, and +360**
- **Monocytopenia (<.2 × 10^9/L) at days +30, +90, +180, and +360**
- **Severe (grade 3-4) aGVHD: yes vs no‡**
- **Chronic GVHD: no vs limited vs extensive**

Continuous variables were first analyzed and then reanalyzed as qualitative variables if a statistical trend was found for any outcome.

*Reduced intensity conditioning regimens were used only in BMT and PBSCT recipients; hence, this variable was not used in the comparison of CBT with BMT/PBSCT groups, but only in the outcome analyses of these latter transplantation groups.

†Second-transplant and advanced disease status were strongly associated (or showed strong collinearity) with each other; thus, only the variable that showed $P < .01$ and had the highest hazard ratio in univariate testing was included in multivariate models if both variables were identified as risk factors in univariate analysis.

‡All of these variables were analyzed as time-dependent covariates.

§Use of high-dose steroids after transplantation and ATG were strongly associated (or showed strong collinearity) with the development of severe (grade 3-4) aGVHD; thus, only the latter was included in multivariate models, because it showed the highest hazard ratio with P value <.01 in all outcomes for which they were identified as risk factors in univariate analysis.

groups were combined for univariate and multivariate analyses, although they are tabulated separately in the descriptive Tables 1, 2, 3 and 5.

RESULTS

Hematologic Recovery and GVHD

Detailed characteristics of hematologic recovery and GVHD by stem cell transplant source are listed in Table 1. Median times to reach a neutrophil count of .5 × 10^9/L, a monocyte count of .2 × 10^9/L, and a lymphocyte count of .2 × 10^9/L were shorter in the BMT/PBSCT groups. There was a trend for a lower incidence of severe aGVHD in the CBT group (18% versus 24%, $P = .1$). A similar trend was found in the 3-year cumulative incidence of extensive cGVHD ($P = .1$).

Severe Infections

A detailed description of all severe infections is presented in Table 2. Two hundred ninety-eight severe infections were documented in 140 of 192 patients. Frequencies of severe infections at 3-year follow-up were 41 of 48 (85%), 83 of 120 (69%), and 16 of 24 (67%) in recipients of CBT, BMT, and PBSCT, respectively ($P = .009$ for CBT group versus BMT/PBSCT group). There was a trend for a higher incidence of any severe infection during the early period (before day +30) in the CBT group (54% versus 25%, $P = .03$), whereas incidences were similar between groups in the intermediate, late, and very late periods (30%, 12%, and 15%, respectively, for CBT recipients and 35%, 20%, and 19%, respectively, for BMT/PBSCT recipients).

Fifty-six patients (29%) had ≥2 severe infectious episodes from different pathogens, without differences between groups. In the early-intermediate periods after transplantation (before day + 100), bacterial infections accounted for most severe infections (55% of episodes versus 14% IFIs and 32% viral infections) as opposed to the late/very late periods, when viral infections represented 55% of severe infectious complications ($P < .01$). These differences in pathogen types between periods were similar among stem cell source transplantation groups (Table 2).

Infection-Related Mortality

As presented in Table 3, IRM was the primary or secondary cause of death (with or without another major cause, mostly GVHD, respectively) in

Table 5. *Summary of Results of Multivariate Analyses for IRM, CMV Infection, and NRM**

	Overall Analysis		CBT (n = 48)		BMT+PBSCT (n = 144)	
	Multivariate *P*	HR (95% CI)	Multivariate *P*	HR (95% CI)	Multivariate *P*	HR (95% CI)
IRM (n = 72)*						
Cord blood transplant as stem cell source	.5	—				
Neutropenia (<0.2×10⁹/l) on day +30†	<.01	3.9 (2.3-4.8)	.05	NR	.04	NR
Monocytopenia on day +30 or at the start of each period†	<.001	4.7 (1.9-16)			<.01	2.6 (1.2-14)
Acute GVHD grades 3-4†	.01	2.4 (1.2-4.7)			<.001	3.5 (2-4.2)
Low stem cell dose infused‡	<.01	2.9 (1.2-3)	.05	NR	.08	NR
Any HLA mismatch	.06§	NR	.7	NR	.05‖	
CMV infection (n = 75)						
Any HLA mismatch	<.05§	NR	.5		.08‖	
Acute GVHD grades 3-4†	<.001	2.7 (1.7-4.4)			<.001	3.6 (2-6)
Patient CMV immunoglobulin G positive	<.001	4.3 (2.3-oo)	.02	2.5 (1.6-oo)	<.0001	4.8 (2.2-oo)
Cord blood transplant as stem cell source	.2	—	—	—		
Extensive chronic GVHD†					.05	NR
NRM						
Cord blood transplant as stem cell source	.4	—				
Second HSCT	.03	NR			<.01	2.2 (1.2-4)
Monocytopenia on day +30†	<.001	4.5 (1.4-18)			<.001	11 (6.5-28)
Acute GVHD grades 3-4†	.01	2.1 (1.2-2.6)	.8#		.01	1.9 (1.2-3.2)
Any HLA mismatch	.07§	NR			.01‖	2.5 (1.7-3.3)
Neutropenia on day +30†			.05	NR		
Low stem cell dose‡	<.01	2.2 (1.6-3.1)	.02	4.1 (2.2-7.1)		
IFIs (n = 38)			Univariate		Univariate	
Early-intermediate periods (before day +100) (IA = 14/IC = 12)			n = 6		n = 19	
Cord blood transplant as stem cell source	.3	—				
Acute GVHD grades 3-4†	.001/NS	4.1 (1.9-15)	.01	—	.001	—
Neutropenia on day +30†	.01/0.03	NR/NR	.01	—	.01	—
Conventional conditioning	Not applicable	—	Not applicable		.04	—
CMV disease†	.07/NS	NR/—			.04	—
Use of ATG for treating aGVHD†	.02/—	NR/—			<.01	—
Monocytopenia on day +30†	.05/NS	—	.05	—	—	—
Late-very late periods (> day +100) (All IA = 12)			No cases		n = 12	
BM-PBSC as stem cell source	.05	—				
Acute GVHD grades 2-4†	.001	6 (2.5-9.5)			.001	
CMV disease†	.09	NR			.05	
Lymphocytopenia on day +90 or at the start of each period†	.08	NR			.02	
Extensive chronic GVHD†	.01	3 (1.3-142)			<.001	
Monocytopenia on days +90 and +180†	—	—			.04	
Use of ATG for treating aGVHD†	—	—			<.01	
CMV disease (n = 22)			Univariate		Univariate	
Cord blood transplant as stem cell source	.3	—				
Prior CMV infection	.04	NR	<.01		.01	
Extensive chronic GVHD†	<.01	9 (2.4-33)	<.001		<.01	
Patient CMV IgG positive	<.001	14 (10-oo)			.001	
Acute GVHD grades 3-4†	—	—			.01	
Conventional conditioning	Not applicable	—			.04	

*For invasive fungal infections and CMV disease, multivariate analysis was done for all 192 patients, but only univariate results for each transplantation group are shown, due to the few events per group. The stem cell transplant group (CBT vs BMT/PBSCT) was introduced in all overall multivariate analysis, despite being nonsignificant in univariate testing, because it was the main objective of this study.

BMT indicates bone marrow transplantation; PBSCT, peripheral blood stem cell transplantation; CBT, cord blood transplantation; GVHD, graft-versus-host disease; CMV, cytomegalovirus; IFI, invasive fungal infection; HSCT, hematopoietic stem cell transplantation; NR, not reported if $P > .01$, as described in Statistical Analysis; HR, hazard ration; CI, confidence interval. HRs and 95% CIs are shown for all significant variables.

Monocytopenia on day +30 or at the start of each period refers to the presence of monocytopenia at the start of each period after transplantation (intermediate, late, and very late).

*Other variables included in the multivariate analysis for IRM in all 192 patients were advanced disease status (univariate $P = .01$) and chronic extensive GVHD (univariate $P < .01$). In the BMT-PBSCT group, the main variable that was nonsignificant in the multivariate analysis was chronic extensive GVHD (univariate $P = .02$). In the CBT group, nonsignificant variables in multivariate analysis were advanced disease status (univariate $P = .06$) and chronic extensive GVHD (univariate $P = .02$).

†Factors entered into the model as time-dependent covariates.

‡Low stem cell dose is defined in text.

§Refers to any HLA mismatches identified by serology (antigen mismatch) or by low-resolution PCR-based mismatches in HLA-A and -B and DRB1 by high-resolution PCR allele-level testing (4-5/6 vs 6/6).

‖Refers to any HLA mismatches identified by serology (antigen mismatch) or by high-resolution allele-level PCR testing in HLA-A, -B, -C, and DRB1 (7/8 vs 8/8), as detailed in text.

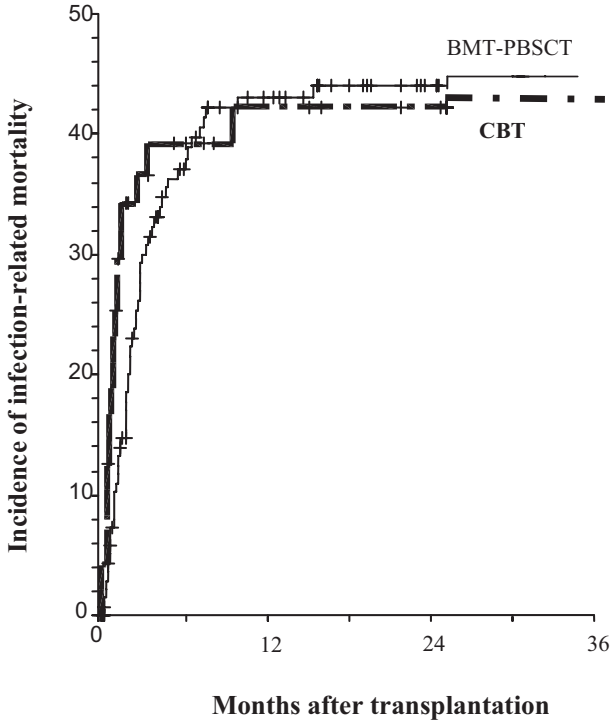

Figure 1. Three-year cumulative incidence of IRM by stem cell transplantation group.

>55% of all deaths in all stem cell source transplantation groups. The 100-day and 3-year incidences of IRM did not differ between groups (Table 3 and Figure 1). Specific causes of death are presented in detail in Table 3, but due to the few cases for most specific pathogens, no statistical comparisons can be done between transplantation groups, although there appeared to be no large differences between them. The proportion of IRMs that occurred during the first 100 days after CBT (14 of 19, 73%) was higher than in the BMT/PBSCT groups (25 of 50, 50%; $P = .02$).

Table 5 presents multivariate analyses of risk factors for IRM in all 192 patients and in each transplantation group studied. An interesting finding is that neutropenia on day +30 and monocytopenia on day +30 or at the last period analyzed had the strongest negative effect on IRM (highest hazard ratio) in multivariate analyses of all 192 patients. The negative effect of monocytopenia was also found in BMT/PBSCT recipients, although neutropenia on day +30 showed a trend in both transplant types. Of note, the presence of any HLA mismatch in BMT/PBSCT recipients showed a trend for a higher IRM (and clearly increased NRM and decreased OS, as described below in Table 5). In contrast, the presence of 0 to 1 mismatch or 2 mismatches in A, B, and DRB1 in CBT recipients had no effect on IRM or any other transplantation outcome.

CMV Infection and Disease

Seventy-five patients (39% overall, 52% in CMV-seropositive patients) presented CMV infection and 22 with CMV disease (detailed in Tables 2 and 5). The 3-year cumulative incidences of CMV infection and disease were similar in all transplantation groups (29% in CBT versus 30% in BMT/PBSCT recipients and 7.2% versus 6%, respectively). Of note, the proportion of CMV disease occurring after day +100 was high in both transplantation groups (4 of 7, 57%, in CBT group and 6 of 15, 40%, in BMT/PBSCT group). Two additional cases of late CMV disease occurred in 2 patients with previous CMV disease before day +100.

However, CMV infection showed a trend for earlier onset in CBT recipients. Median days after HSCT of onset of CMV infection was +52 (range, 9-244) in CBT recipients and +82 (range, 35-859) in BMT/PBSCT recipients ($P = .05$), and proportions of CMV infections that occurred before day +30 was higher in the CBT group (35% versus 7%, respectively; $P = .006$). Quantitative CMV antigenemia was available in 70 of 75 patients with CMV infection. To determine whether there was any correlation between the magnitude of CMV antigenemia and subsequent development of CMV disease, we compared the maximum number of positive cells in patients with and without CMV disease, without any apparent correlation (data not shown).

In univariate and multivariate analyses, patient CMV seropositivity was the major risk factor for CMV infection and disease, and previous CMV infection treated preemptively with ganciclovir and/or foscarnet increased the risk of developing disease (Table 5). More specifically, all 10 cases of a first episode of CMV disease that occurred after day +100 had been successfully treated preemptively for CMV infection before day +100, and 2 additional patients, who had developed an episode of CMV disease before day +100 and had been successfully treated, had a second episode of CMV disease after day +100. Transplantation group had no effect on risk of CMV infection and disease. Nevertheless, when analyzing only seropositive patients for CMV before HSCT, there was a trend for a higher risk of CMV disease in CBT recipients. Other variables included in the multivariate analysis for CMV infection in all 192 patients were use of antithymocyte globulin for treating aGVHD (univariate $P < .05$) and lymphocytopenia ($<.2 \times 10^8$/L on days +30 and/or +90, univariate $P = .03$). In the BMT/PBSCT and CBT groups, the only variable that was nonsignificant in multivariate analysis was lymphocytopenia ($<.2 \times 10^8$/L on days +30 and/or +90, univariate $P = .04$ and .01, respectively).

Because extensive cGVHD was the strongest risk

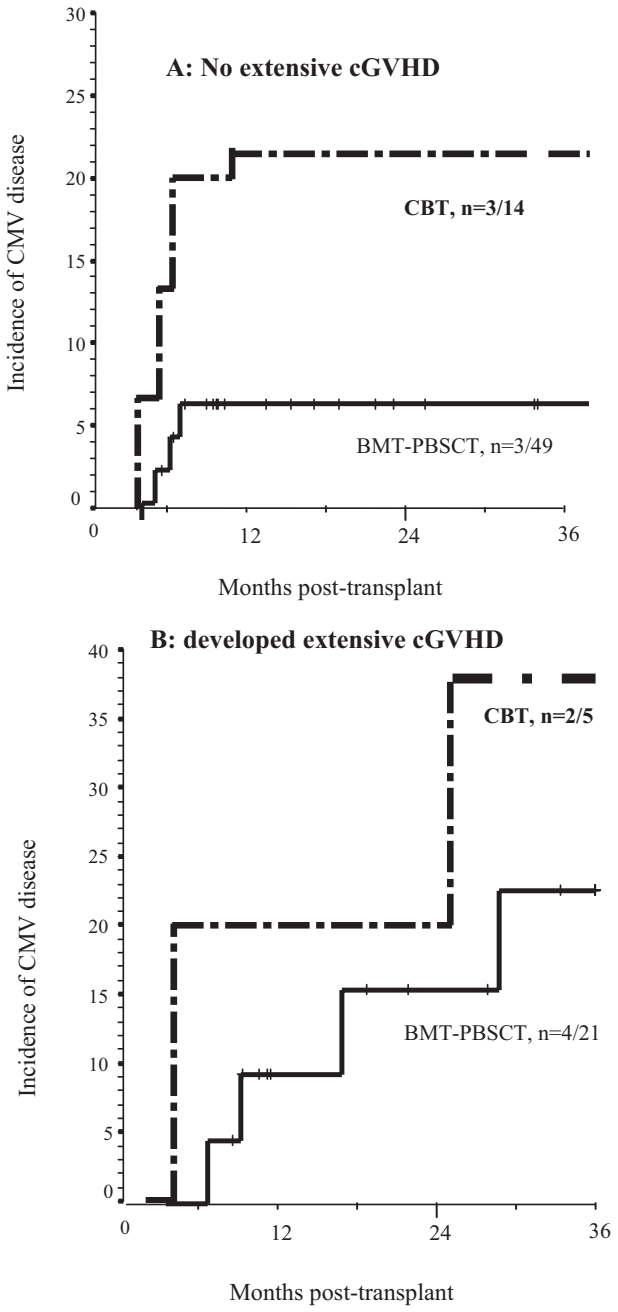

Figure 2. Landmark analysis (including only patients alive and well on day +100 after transplantation) of 3-year cumulative incidence of CMV disease in patients seropositive for CMV before transplantation by (A) not developing or (B) developing extensive cGVHD after day +100, split by stem cell source transplantation group.

factor for CMV disease in both transplantation groups in univariate analysis, we compared the incidence of late CMV disease by transplantation group and extensive cGVHD. For this comparison, a landmark analysis was done, including only seropositive patients who were evaluable for cGVHD (alive and well on day +100; CBT, n = 19; BMT/PBSCT, n = 70), and calculated the incidence of CMV disease in each group. As seen in Figure 2, there was a trend for a higher rate of CMV disease after day +100 in both subgroups of CBT recipients (2 of 5, 40%, with extensive cGVHD and 3 of 14, 21%, without extensive cGVHD) compared with BMT/PBSCT recipients (4 of 21, 19%, and 3 of 49, 6%, respectively). Due to the small numbers of patients and events per subgroup, no statistical comparison was done.

Recurrence of CMV Infection

Sixteen patients (21% of patients with CMV infection) had recurrent CMV infections (11 had 2 episodes and 4 had >2 episodes). Risk factors for recurrent CMV infections could not be analyzed due to the small samples, but the rate was similar in all transplantation groups and recurrent infection apparently did not increase the risk of CMV disease.

Other Severe Viral Infections

Severe infections by all other viruses were diagnosed less commonly than CMV (Table 2). Thirty-nine cases of nonlocalized infections by other herpes viruses occurred, and varicella-zoster virus infections were the most pathogens in all study groups (23% of CBT recipients and 10% of BMT/PBSCT recipients). Most varicella-zoster virus infections occurred after day +100 (11 of 11, 100%, and 11 of 13, 87%, respectively).

Less common non-CMV viral infections included Epstein-Barr virus lymphoproliferative disease and community respiratory virus infections in 3% and 4% of patients, respectively. However, only 2 patients died from non-CMV viral infections (disseminated adenovirus disease and respiratory syncytial virus pneumonitis), although both patients also had CMV disease and IA.

Eleven patients (6.5%) developed hemorrhagic cystitis with the presence of *Polyoma* virus in urine. However, because this observation did not suffice to attribute the cystitis to this virus, we did not include these cases as severe viral infections. All episodes of cystitis resolved with supportive measures.

Bacterial Infections and Pneumonia of Unknown Origin

Seventy-two patients (37%) presented 108 severe bacterial infections after transplantation. All bacterial species found are shown by period and type of stem cell source in Table 2. Gram-negative bacilli infections predominated in all periods and stem cell transplantation groups, but this predominance of gram-negative bacilli was due to exclusion of coagulase-negative staphylococci from analysis, which account for most infections in all recent large studies [2,4,9,29].

Although there were no relevant differences in any specific bacterial pathogens found, CBT recipients had a higher rate of any serious bacterial infection

before day +100. Thus, the bacterial infection density before day +100 in CBT recipients was higher than that in BMT/PBSCT recipients (21 infections per 1000 patient-days in CBT group versus 14 in BMT/PBSCT group, respectively; $P = .04$). Twenty-two patients died from a bacterial infection, as shown in Table 5. The highest mortality was seen for bacterial pneumonias (14 of 23 episodes, 61%) and meningitis (4 of 4 episodes).

Forty-one patients (21%) had ≥1 episode of clinically defined pulmonary infection but no pathogen was identified (pneumonia of unknown origin or PUO), with similar rates in all transplantation groups and similar attributable mortality (5 of 7 in CBT group and 22 of 34 in BMT/PBSCT group, $P = .8$). At day +100 and at +3 years the cumulative incidences of PUO were 7% and 15%, respectively, in CBT recipients and 10% and 17% in BMT/PBSCT recipients ($P = .3$). In the CBT group, 3 of 7 cases of PUO (43%) occurred before day +100 and 4 of 7 (57%) occurred >1 year after transplantation; in the BMT/PBSCT group, the time distributions were 21 of 34 (62%) and 12 of 34 (35%), respectively ($P = .7$). In univariate analysis, the only risk factors that showed a trend for PUO were advanced disease status ($P = .07$), mismatched sex ($P = .06$), and the presence of lymphocytopenia on day +30 ($P = .03$).

Invasive Fungal Infections

Thirty-eight patients (19%) developed 40 episodes of a probable or proved IFI. Twenty-six episodes (71%) were IA, and 12 (29%) were caused by non-*Candida albicans* (see Table 2 for details). There was 1 episode of *P jiroveci* pneumonia, 1 of *Penicillium marneffei* fungemia, and 1 of histologically proved mucormycosis. Death from an IFI did not differ between transplantation groups (22 of 26 cases of IA, 85%, and 5 of 12 cases of invasive candidiasis, 42%).

The CBT group showed a statistically significant trend ($P < .01$) in developing a higher proportion of IFIs before day +100 (12 of 12 IFIs) compared with the other group (11 of 25, 40%). However, the 3-year cumulative incidence of developing an IFI in all 192 patients was 12% (95% confidence interval [CI], 6 to 18) and did not differ between transplantation groups. Incidences of IA and candidiasis were 10% and 2%, respectively. Median times to onset were 148 days (range, 14-622) for IA and day +48 (range, 4-122) for candidiasis.

To estimate possible risk factors for IA, only univariate analyses are reliable, and the results are shown in detail in Table 5. Before day +100, development of severe (grades 3 to 4) aGVHD and delayed neutrophil recovery increased the risk of IA in CBT and BMT/PBSCT recipients. Risk factors for *late* cases of IA (after day + 100) included aGVHD grades 2 to 4, extensive cGVHD, and use of antithymocyte globulin for treating severe GVHD. Of note, stem cell source had no effect in early IA, although a trend for a higher risk in the BMT/PBSCT group was found (12 of 95 evaluable patients, 13%, versus 0 of 28 CBT recipients, respectively; $P = .05$). No risk factors were found for the 12 cases of *Candida* infection, except for a trend for neutropenia on day +30. Interestingly, all cases were due to non–*Candida albicans* species.

Other Severe Infections and Simultaneous Infections

Numerous uncommon opportunistic infections were found, as detailed in Table 2. Of note, toxoplasmosis disease was diagnosed before death in 4 cases (2 CBT and 2 PBSCT recipients), but the actual frequency in patients at risk (patients seropositive for immunoglobulin G for *Toxoplasma gondii* before transplantation) was 4 of 77 (5%), with 3 patients dying from disseminated toxoplasmosis.

Twenty-one patients (11%) had ≥1 episode of simultaneous severe infections, which had a high attributable mortality (76%). The most common associations were fungal and viral infections. CMV pneumonia preceded or occurred simultaneously with probable or proved aspergillosis in 7 cases (27% of IA), and 2 cases of IA coexisted with an Epstein-Barr virus lymphoproliferative disease and respiratory syncytial virus pneumonia, respectively.

NRM and OS

NRM occurred in 20 (41%) and 76 (52%) patients in the CBT and BMT/PBSCT groups, respectively, at medians of 149 days (range, 16-573) and 189 days (range, 69-671) after transplantation, respectively. The 100-day and 3-year cumulative incidences of NRM in the CBT versus BMT/PBSCT group were 15% (95% CI, 7 to 39) versus 12% (95% CI, 7 to 25) and 48% (95% CI, 13 to 99) versus 41% (95% CI, 29 to 49), respectively ($P > .2$ for both comparisons). The specific causes of NRM and death and univariate and multivariate analyses of risk factors for NRM are presented in Table 5.

Thirty-nine percent of all patients were alive at final follow-up, with a median follow-up of 50 months (range, 24-95). OS at 3 years was 36% in the CBT group and 39% in the BMT/PBSCT group ($P = .7$). In multivariate analysis by stem cell source group, variables that decreased 3-year OS in the BMT/PBSCT group were (1) a previously failed autologous HSCT ($P = .004$), (2) monocytopenia on day +30 ($P = .01$), (3) grade 3/4 aGVHD ($P = .01$), (4) not developing limited cGVHD ($P = .0001$), and (5) any HLA mismatch by high-resolution typing ($P < .01$). In the CBT group, deleterious variables for OS were (1) age >35 years ($P < .01$), (2) advanced disease status

at transplantation ($P = .02$), (3) neutropenia on day +30 ($P = .02$), and (4) low nucleated cell dose infusion ($P < .01$).

DISCUSSION

Most single-center and multicenter or registry-based studies have shown that adult recipients of a VUD HSCT have a high risk of developing severe infections, although most studies to date are hampered by the description of selected homogeneous patient cohorts, by the lack of specific data on the true HLA mismatches between donor and recipients, and by a lack of comparison of CBT as a source of stem cells [1,2,10,27-30]. Clinicians who perform VUD transplantation in adults are aware that severe infections are very common, generate important morbidity, and require costly and complex diagnostic and therapeutic interventions. Despite the obvious limitations of the present multicenter and retrospective study, we aimed at describing the exact numbers and risk factors for severe infections in all VUD transplantations performed during a recent period, without patient selection, in an effort to describe the problems faced in real clinical practice, However, the statistical analysis was rigorously designed (detailed in Statistical Analysis), so comparative analyses of outcomes were more reliable; nevertheless, undetected biases can lead to incorrect observations in any retrospective study.

Our findings on the frequencies and cumulative incidences of major life-threatening infections are in line with previous studies in VUD HSCT in adults, mainly IFIs (13% cumulative incidence), CMV infection and disease, and IRM [4,5,10,11]. With respect to the 2 former severe infections, we found that many cases occurred late after transplantation (after day +100), as reported recently [4,5,10,30]. Thus, 54% of IFIs and 45% of CMV disease occurred after day +100, with similar proportions in all transplantation groups. In addition, >30% of IRM occurred after day +100. Although the definitions may vary somewhat, IRM was responsible for >50% of all cases of NRM in other studies of VUD HSCT in adults [13-15,31-33].

Because CBT has been investigated as a source of alternative donor stem cell source for VUD transplantation in adults only recently, there is little detailed information on the infectious complications after CBT, especially when compared with other VUD stem cell sources. Nevertheless, recently published analyses have indicated that survival appears to be similar to BMT from VUD [12-14]. The patterns and types of severe infections seen in CBT in adults are important to clinicians, because the feasibility of CBT in adults was questioned by many physicians only a few years ago, due to the anticipated high risk of graft failure, delayed hematopoietic recovery because of the small number of stem cells infused, and lack of immune reconstitution [10,12,13,15,29,34].

Our study shows that CBT recipients have a higher incidence of severe infections, especially bacterial infections, in the first 100 days after transplantation. In addition, the proportions of CMV infections, IFIs, and IRMs that occurred before day +100 were higher in CBT recipients than in BMT/PBSCT recipients. However, after day +100, CBT recipients appeared to have a lower risk of most severe infections. Most importantly, the 100-day and 3-year incidences of IRM and NRM were not different between groups. In any type of allogeneic HSCT, the incidence and risk factors for bacterial, fungal, and viral infections are correlated with the kinetics of myeloid and immune reconstitution [29,30,35]. Early infections are mainly attributed to neutropenia and to the damage of mucosal barriers, and the higher risk of early infections may reflect the delay in myeloid recovery in CBT with respect to BMT/PBSCT recipients [11,16,17]. Several ongoing studies are attempting to fasten hematopoietic recovery and thereby decrease infection in the early period after CBT, such as coinfusion of a third-party, haploidentical, fully T-cell–depleted PBSCT from a haploidentical relative [36], and a comparison of early infections with and without such strategies would be of great interest.

After engraftment, cell-mediated immunity is of utmost importance, and infections are increased in case of GVHD and its immunosuppressive treatment [37,38]. Thus, we made an effort to analyze the effect on postengraftment infections of neutrophil, lymphocyte, and monocyte counts as time-dependent covariates. An interesting observation was the strong effect of monocytopenia in multivariate analyses on NRM and IRM (especially in the BMT/PBSCT group) and on IFIs. In support of our findings, a study by Storek et al [39] in Seattle found that a low monocyte count on day +80 was an independent risk factor for the development of any severe infection and severe viral and fungal infections after day +100 in 103 BMT recipients. In addition, a recent abstract from a multicenter 1-year study in France found that monocytopenia was an independent risk factor for a poor outcome of IA among 51 HSCT recipients [40]. The exact reason for these observations is unclear, and the monocyte count could simply be a surrogate marker of the truly relevant biologic mechanism of its protective effect or could have a direct role on nonspecific immune defense mechanisms [39]. As expected, we found that severe GVHD increased the risk of most severe infections before and after day +100. The lower risk of severe cGVHD seen in CBT [12,13] may explain the trend for lower risk of late infections, especially IFIs, seen in our study.

Rocha et al [13] found a significantly lower NRM and overall mortality after HLA-matched VUD BMT but similar outcomes between CBT and HLA-mismatched VUD BMT. Our results are in line with this study, because we found a lower IRM in 8of 8 allele-matched BMT/PBSCT recipients with respect to mismatched BMT/PBSCT and CBT recipients ($P < .01$), whereas the level of HLA matching had no effect on any outcome in CBT recipients [12,15]. In addition, a high cell dose was a protective variable for IRM and NRM in CBT recipients, thus emphasizing the need of using the cellularity of the cord blood unit as the main variable when selecting a unit for transplantation [15]. However, our results must not be overinterpreted, because the small number of cases at each level of mismatching gives the study low statistical power to detect an effect of the number of mismatches. Moreover, the definition of an HLA match in this study included only 6 HLA loci and low-resolution typing for class I antigens in CBT recipients, whereas high-resolution matching was available for a large proportion of BMT/PBSCT recipients. This handicap was expected, because, in accord with current recommendations, cord blood units were typed with low resolution for HLA-A and HLA-B during the study period, but BMT/PBSCT donors were usually selected with the use of criteria that included high-resolution typing for 8 or 10 HLA antigens [41-43]. However, most experts agree that, when large numbers of CBT have been typed by high-resolution HLA typing, mismatches that lead to worse outcomes, including IRM, may be identified [14,41,44].

In conclusion, with a median follow-up of >3 years, unrelated donor CBT in adults was associated with similar risks of IRM and all major severe infections with respect to VUD BMT/PBSCT. These results highlight that, with an appropriate selection of cord blood units, CBT should continue to be developed as a source of alternative donor HSCT in patients who lack an allele-level matched adult donor. The true relevance of monocytopenia as an independent risk factor for some severe infections merits further studies.

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