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ORIGINAL ARTICLE

Impact of graft-versus-host disease after reduced-intensity conditioning allogeneic stem cell transplantation for acute myeloid leukemia: a report from the Acute Leukemia Working Party of the European group for blood and marrow transplantation

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This report investigated the impact of graft-versus-host disease (GVHD) on transplantation outcomes in 1859 acute myeloid leukemia patients given allogeneic peripheral blood stem cells after reduced-intensity conditioning (RIC allo-SCT). Grade I acute GVHD was associated with a lower risk of relapse (hazards ratio (HR) = 0.7, $P = 0.02$) translating into a trend for better overall survival (OS; HR = 1.3; $P = 0.07$). Grade II acute GVHD had no net impact on OS, while grade III–IV acute GVHD was associated with a worse OS (HR = 0.4, $P < 0.0.001$) owing to high risk of nonrelapse mortality (NRM; HR = 5.2, $P < 0.0001$). In time-dependent multivariate Cox analyses, limited chronic GVHD tended to be associated with a lower risk of relapse (HR = 0.72; $P = 0.07$) translating into a better OS (HR = 1.8; $P < 0.001$), while extensive chronic GVHD was associated with a lower risk of relapse (HR = 0.65; $P = 0.02$) but also with higher NRM (HR = 3.5; $P < 0.001$) and thus had no net impact on OS. *In-vivo* T-cell depletion with antithymocyte globulin (ATG) or alemtuzumab was successful at preventing extensive chronic GVHD ($P < 0.001$), but without improving OS for ATG and even with worsening OS for alemtuzumab (HR = 0.65; $P = 0.001$). These results highlight the role of the immune-mediated graft-versus-leukemia effect in the RIC allo-SCT setting, but also the need for improving the prevention and treatment of severe GVHD.

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Keywords: reduced-intensity conditioning; AML; GVHD; chronic; graft-versus-leukemia effects

INTRODUCTION

Allogeneic hematopoietic stem cell transplantation (allo-SCT) following myeloablative conditioning regimen is the treatment of choice for many young patients with acute myeloid leukemia (AML).[1,2] The antileukemic activity of myeloablative allo-SCT relies not only on the high dose of chemo/radiotherapy given during the conditioning regimen, but also on the immune-mediated graft-versus-leukemia (GVL) effect.[1,3,4] The biology of the GVL effect has been thought to involve reactions to polymorphic minor histocompatibility antigens expressed either specifically on hematopoietic cells (and thus not causing graft-versus-host disease (GVHD)) or more widely on a number of tissue cells.[5,6] Allo-SCT following reduced-intensity conditioning (RIC) is being increasingly used as treatment for patients with AML who are too old or too frail to tolerate high-dose conditioning regimens.[7–19] The goal of RIC allo-SCT is to harness the GVL effect,[7] while minimizing toxicities and the risk of GVHD. However, this is a delicate balance as a number of prior studies have shown a lower risk of relapse in AML patients who experienced chronic GVHD after RIC allo-SCT compared with those patients who did not,[10–12,20,21] while some other studies failed to find such an association.[22,23] A recent analysis from the Center for International Blood and Marrow Transplant Research (CIBMTR) analyzed the impact of GVHD in a cohort of patients with AML or myelodysplastic syndrome given various graft types after RIC conditioning.[24] In a landmark analysis at 1 year after transplantation ($n = 630$), relapse incidence was reduced only in the group of patients with prior both acute and chronic GVHD, while acute and/or chronic GVHD had no significant impact on disease-free survival. With this background, the current report investigated the impact of occurrence of GVHD and of GVHD severity on transplantation outcomes in a large cohort of AML patients given allogeneic peripheral blood stem cells (PBSC) after RIC.

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The study has been presented in part as an oral abstract at the 53rd Annual Meeting of the American Society of Hematology, 10–13 December 2011, San Diego, CA, as well as in the 2012 meeting of the European group for Blood and Marrow Transplantation, Geneva 1-4.

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PATIENTS AND METHODS

Data collection

This was a retrospective study performed by the Acute Leukemia Working Party (ALWP) of the European group for Blood and Marrow Transplantation (EBMT) group. EBMT registry is a voluntary working group of more than 500 transplant centers, participants of which are required to report all consecutive stem cell transplantations and follow-up once a year. The scientific board of the ALWP of EBMT approved this study. Data of adult AML patients in first or second complete remission (CR) at transplantation and given G-CSF-mobilized PBSCs from HLA-identical sibling (MSD) or HLA-matched unrelated donors (MUD) between 2000 and 2009 after RIC were included. Patients given *ex-vivo* T-cell depleted grafts, those given other stem cell sources than PBSC, those given pre-emptive donor lymphocyte infusions and those who failed to engraft (defined as failure to achieve a sustained engraftment of absolute neutrophil count of higher than 0.5×10^9/l; $n = 34$) were excluded. The date and severity (limited versus extensive) of chronic GVHD (graded according to established criteria[25]) were prospectively collected using the EBMT Mimimum Essential Data-A form.

Patients and conditioning

Data from 1859 patients given PBSC from MSD ($n = 1208$) or MUD ($n = 651$) were included. Patients' characteristics are summarized in Table 1. Briefly, median patient age was 56.3 (range, 18–77) years. Disease status at allo-SCT was first CR in 1439 (77%) patients, while the remaining 420 patients were in second CR. The RIC regimen was based on low-dose total body irradiation in 520 (28%) patients, while the remaining patients received chemotherapy-only-based RIC.

Statistical analysis

Cumulative incidence curves were used for relapse incidence and nonrelapse mortality (NRM) in a competing risk setting, as death and relapse were competing together.[26] For estimating the cumulative incidence of GVHD, death was considered as a competing event. Overall (OS) and leukemia-free (LFS) survivals (starting from date of transplant) were calculated using the Kaplan–Meier estimates; the log-rank test was used for univariate comparisons. Associations of patient and graft characteristics with grade II–IV acute GVHD were evaluated using multivariate logistic regression. Associations of patient and graft characteristics with other outcomes (chronic GVHD, relapse, NRM, LFS and OS) were evaluated in multivariable analyses, using Cox proportional hazards. Factors included in the models for acute and chronic GVHD included donor type, patient age >56 years, female donor to male recipient versus other gender combinations, donor and recipient cytomegalovirus serostatus, disease status at the time of transplant, cytogenetic risk group, TBI- versus chemotherapy-based RIC, the use of antithymocyte globulin (ATG) in the conditioning, the use of alemtuzumab in the conditioning, center activity[27] (arbitrarily defined as center that performed $\leqslant$or $\geqslant$20 first allo-SCT with RIC conditioning as treatment for AML between 2000 and 2009) and prior grade II–IV acute GVHD (for chronic GVHD). The same factors as well as acute, limited chronic and extensive chronic GVHD were included in the models for NRM, relapse, LFS and OS (Table 2). The impact of chronic GVHD on other outcomes was always studied as a time-dependent variable, and we only considered GVHD occurring before relapse in the paper. All tests were two sided. The type I error rate was fixed at 0.05 for determination of factors associated with time to event outcomes. Statistical analyses were performed with SPSS (SPSS Inc., Chicago, IL, USA) and Splus software (MathSoft Inc., Seattle, WA, USA) packages.

RESULTS

Transplantation outcomes

Acute GVHD of grades II, III and IV were observed in 252 (14%), 102 (5%) and 54 (3%) patients, respectively, while grade unknown, limited and extensive chronic GVHD were observed in 30 (2% of patients alive at day 100 ($n = 1545$)), 349 (23%) and 391 (25%) patients, respectively (Table 1). With a median follow-up of 28 (range, 0.5–130) months, 3-year incidences of relapse, NRM, LFS and OS were $34 \pm 1\%$, $18 \pm 1\%$, $48 \pm 1\%$ and $54 \pm 1\%$, respectively. Supplementary Table S1 shows the factors associated with outcomes in univariate analyses.

Table 1. Patients and transplant outcomes

	N = *1859*
Median patient age, years (range)	56.3 (18–77)
Recipient gender (M/F), # M (%)/#F (%)	946 (51)/912 (49)
Donor type # MRD (%)/ # MUD[a] (%)	1208 (65)/ 651 (35)
Median donor age (range), years	48.6 (12–80)
Donor gender (M/F), #M (%)/#F (%)	1101 (60)/ 742 (40)
Female donor/male recipient, # pts (%)	338 (18)
Median year of SCT, years (range)	2006 (2000–2009)
Status at SCT # CR1 (%)/ # CR2 (%)	1439 (77)/ 420 (23)
Patient CMV seropositivity # (%)	1051 (66)
Donor CMV seropositivity # (%)	885 (56)
Cytogenetics	
Good risk[b]	88 (5)
Intermediate risk[c]	776 (42)
High risk[d]	192 (10)
Not reported	803 (43)
Conditioning (RIC[e]), # (%)	
Low-dose TBI-based RIC	520 (28)
Fludarabine-melphalan-based RIC	390 (21)
Fludarabine-busulfan-based RIC	654 (35)
Other chemotherapy-based RIC	295 (16)
In-vivo *T-cell depletion, # (%)*	
No	1007 (54)
ATG	536 (29)
Alemtuzumab	316 (17)
Center activity, # (%)	
Higher[f]	1408
Lower	451
Grade II–IV acute GVHD, # (%)	408 (22)
Chronic GVHD	
Limited chronic GVHD, # (%[g])	349 (23)
Extensive chronic GVHD, # (%[g])	391 (25)
Chronic GVHD grade unknown, # (%[g])	30 (2)
3-year CI of chronic GVHD	47 ± 1
Median (range) time of diagnosis of chronic GVHD (days)	163 (100–1545)
3-year CI of NRM[h] (%)	18 ± 1
3-year CI of relapse[i] (%)	34 ± 1
3-year LFS[j] (%)	48 ± 1
3-year OS (%)	54 ± 1

Abbreviations: ATG, antithymocyte globulin; CI, cumulative incidence; CMV, cytomegalovirus; CR1, first complete remission; CR2, second complete remission; F, female; GVHD, graft-versus-host disease; LFS, leukemia-free survival; M, male; MRD, HLA-identical related donor; MUD, HLA-matched unrelated donor; NRM, nonrelapse mortality; OS, overall survival; RIC, reduced-intensity-conditioning; SCT, stem cell transplantation; TBI, total body irradiation; #, number of patients. [a]Defined as 6/6 or 8/8 HLA-allele-matched unrelated donors. [b]Defined as t(8;21), t(15;17), inv or del (16), or acute promyelocytic leukemia, these abnormalities only or combined with others. [c]Defined as all cytogenetics not belonging to the good or high risk (including trisomias). [d]Defined as 11q23 abnormalities, complex caryotype, and abnormalities of chromosomes 5 and 7. [e]Defined as use of fludarabine associated with fewer than 6 Gy (low dose) total-body irradiation, or busulfan $\leqslant 8$ mg/kg, or other nonmyeloablative drugs. [f]Defined as center that performed $\geqslant 20$ first allo-SCT with RIC conditioning as treatment for acute myeloid leukemia (AML) between 2000 and 2009. [g]Among patients alive at day 100. [h]Defined as any death without previous relapse or progression. [i]Defined as hematologic relapse. [j]Defined as survival without evidence of relapse.

Impact of GVHD on transplantation outcomes

Predictive factors for grade II–IV acute GVHD included unrelated donor (RR = 2.4; 95% CI, 1.8–3.3; $P < 0.001$), intermediate (RR = 2.7; 95% CI, 1.3–5.6; $P = 0.009$) or poor risk (RR = 2.3; 95% CI, 1.0–5.1; $P = 0.04$)

Table 2. Multivariate analyses of transplant outcomes

	Progression/relapse		*NRM*		*LFS*		*OS*	
	HR[a] *(95% CI)*	P[a]	*HR*[a] *(95% CI)*	P[a]	*HR*[a] *(95% CI)*	P[a]	*HR*[a] *(95% CI)*	P[a]
No chronic GVHD[b] (reference)	1		1		1		1	
Limited chronic GVHD[b]	0.7 (0.5–1.0)	0.07	1.5 (0.9–2.3)	0.4	1.1 (0.9–1.5)	0.42	**1.8 (1.3–2.4)**	**<0.001**
Extensive chronic GVHD[b]	**0.7 (0.5–0.9)**	**0.02**	**3.5 (2.4–5.2)**	**<0.001**	**0.8 (0.6–1)**	**0.03**	**1.1 (0.8–1.4)**	**0.58**
Acute GVHD								
No acute GVHD (reference)	1		1		1		1	
Acute GVHD grade I	**0.7 (0.5–0.9)**	**0.02**	0.9 (0.6–1.3)	0.54	**1.3 (1.1–1.7)**	**0.01**	1.3 (1–1.6)	0.06
Acute GVHD grade II	0.7 (0.6–1)	0.05	1.3 (0.9–1.2)	0.19	1.1 (0.9–1.4)	0.36	1 (0.8–1.2)	0.85
Acute GVHD grade III–IV	0.6 (0.4–1)	0.05	**5.2 (3.7–7.2)**	**<0.001**	**0.5 (0.4–0.7)**	**<0.001**	**0.4 (0.3–0.5)**	**<0.001**
Use of alemtuzumab versus not	**1.5 (1.1–2.1)**	**0.008**	1.4 (1–2.2)	0.08	**0.7 (0.5–0.8)**	**0.001**	**0.6 (0.5–0.8)**	**0.001**
Use of ATG versus not	**1.5 (1.2–2.0)**	**0.008**	0.7 (0.5–1.0)	0.08	0.8 (0.7–1)	0.11	0.9 (0.7–1.2)	0.51
MUD versus MRD	0.9 (0.7–1.1)	0.3	**1.94 (1.3–2.7)**	**<0.001**	0.9 (0.7–1.1)	0.16	0.8 (0.7–1)	0.11
CR2 versus CR1	**1.6 (1.3–2.0)**	**<0.001**	**1.6 (1.2–2.2)**	**0.002**	**0.6 (0.5–0.7)**	**< 0.001**	**0.6 (0.5–0.8)**	**<0.001**
Patient age >56 years	1.1 (0.9–1.4)	0.19	**1.3 (1.0–1.7)**	**0.04**	**0.8 (0.7–1)**	**0.03**	**0.8 (0.7–0.9)**	**0.01**
Female donor to male recipient	0.9 (0.7–1.2)	0.62	1.3 (1.0–1.9)	0.08	1 (0.8–1.2)	0.73	0.9 (0.8–1.2)	0.63
Patient CMV seropositivity	0.9 (0.7–1.1)	0.4	1.1 (0.8–1.5)	0.45	1 (0.9–1.2)	0.97	1.0 (0.8–1.2)	0.98
Donor CMV seropositivity	1.1 (0.9–1.3)	0.67	1.1 (0.8–1.5)	0.48	0.9 (0.8–1.1)	0.43	0.9 (0.8–1.1)	0.40
TBI-based RIC versus not	**1.6 (1.2–2)**	**<0.001**	**0.9 (0.6–1.3)**	0.52	**0.8 (0.6–0.9)**	**0.01**	**0.8 (0.6–1)**	**0.02**
Cytogenetics								
Good (reference)	1		1		1		1	
Intermediate versus good	**2.3 (1.3–3.9)**	**0.003**	1.4 (0.8–2.5)	0.29	**0.5 (0.4–0.8)**	**0.003**	**0.6 (0.4–0.9)**	**0.01**
Poor versus good	**3.6 (2.0–6.4)**	**<0.001**	1.23 (0.7–2.8)	0.42	**0.4 (0.3–0.6)**	**<0.001**	**0.4 (0.3–0.7)**	**0.001**
NA versus good	**2.2 (1.3–3.8)**	**0.005**	1.7 (0.9–3.2)	0.09	**0.5 (0.3–0.8)**	**0.002**	**0.5 (0.3–0.8)**	**0.004**
Big centers[c]	**0.7 (0.6–0.9)**	**0.01**	**0.7 (0.5–1.0)**	**0.03**	**1.3 (1.1–1.6)**	**0.002**	**1.3 (1.1–1.5)**	**0.03**

Abbreviations: ATG, antithymocyte globulin; CMV, cytomegalovirus; CR1, first complete remission; CR2, second complete remission; GVHD, graft-versus-host disease; HR, hazards ratio; LFS, leukemia-free survival; MRD, HLA-identical related donor; MUD, HLA-matched unrelated donor; NA, not applicable; NRM, nonrelapse mortality; OS, overall survival; RIC, reduced-intensity-conditioning; TBI, total body irradiation. [a]Statistically significant factors are in bold. [b]Modeled as a time-dependent event. [c]Arbitrarily defined as a center that performed ⩾20 first allo-SCT with RIC conditioning as treatment for AML between 2000 and 2009.

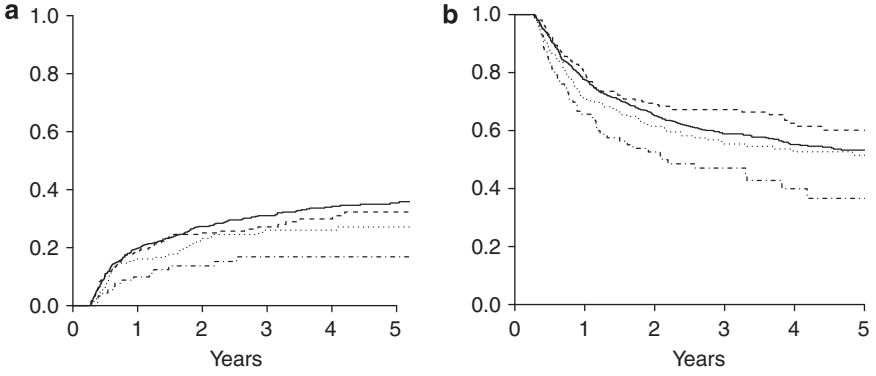

Figure 1. (**a**) Landmark analysis at 100 days after transplantation showing the cumulative incidence of relapse in patients who had no acute GVHD (continuous line) (at 4 years = 34 ± 2%), grade I acute GVHD (broken line — —) (at 4 years = 30 ± 3%), grade II acute GVHD (dotted line - - -) (at 4 years = 26 ± 3%), grade III–IV acute GVHD (broken line – - – - –) (at 4 years = 17 ± 4%). (**b**) Landmark analysis at 100 days after transplantation showing OS in patients who had no acute GVHD (continuous line) (at 4 years = 60 ± 2%), grade I acute GVHD (broken line — —) (at 4 years = 66 ± 2%), grade II acute GVHD (dotted line - - -) (at 4 years = 56 ± 4%), grade III–IV acute GVHD (broken line — - – - –) (at 4 years = 43 ± 4%).

cytogenetics, and absence of *in-vivo* T-cell depletion with alemtuzumab (RR = 0.5; 95% CI, 0.4–0.8; $P = 0.004$). There was also a suggestion for lower risk of grade II–IV acute GVHD in patients given ATG (RR = 0.8; 95% CI, 0.5–1.1; $P = 0.10$), although it did not reach statistical significance. In multivariate analysis, grade I acute GVHD was associated with a lower risk of relapse (hazards ratio (HR) = 0.7, 95% CI, 0.5–0.9; $P = 0.02$) translating into better LFS (HR = 1.3, 95% CI, 1.1–1.7; $P = 0.01$) and a trend for better OS (HR = 1.3, 95% CI, 1.0–1.6; $P = 0.06$). Grade II and grade III–IV acute GVHD were each associated with a lower risk of relapse (HR = 0.7, 95% CI, 0.6–1.0; $P = 0.05$; and HR = 0.6, 95% CI, 0.4–1.0, $P = 0.05$, respectively), but also with a suggestion for higher NRM for grade II acute GVHD (HR = 1.3, 95% CI, 0.9–1.2; $P = 0.19$), and higher NRM for grade III–IV acute GVHD (HR = 5.2, 95% CI, 3.7–7.2; $P < 0.0001$, respectively) leading a similar OS for grade II acute GVHD (HR = 1.0, 95% CI, 0.8–1.2; $P = 0.85$) and a worse OS for grade III–IV (HR = 0.4, 95% CI, 0.3–0.5; $P < 0.001$) acute GVHD. To further assess the GVL effect of acute GVHD, we also performed a landmark analysis in patients who were leukemia-free at 100 days after transplantation. As shown in the Figure 1, 4-year OS were

66 ± 2% in patients with grade I acute GVHD, 60 ± 2% in patients without acute GVHD, 56 ± 4% in patients with grade II acute GVHD and 43 ± 4% in patients with grade III–IV acute GVHD.

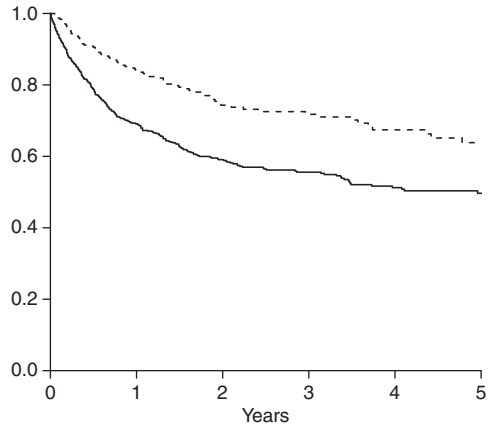

Figure 2. 'Pseudo landmark analysis'[20,29] illustrating the impact of limited chronic GVHD on OS in a time-dependent manner. The 'no chronic GVHD line' (continuous line) shows OS from day 166 after RIC-alloSCT (the median day of onset of limited chronic GVHD) for patients who never experienced chronic GVHD. The limited chronic GVHD line (broken line) shows OS from diagnosis of limited chronic GVHD in patients who experienced limited chronic GVHD anytime after transplantation. At 3 years, survival was 72 ± 3% in the limited chronic GVHD group versus 56 ± 2% in the non-GVHD group ($P < 0.0001$).

Predictive factors for extensive chronic GVHD other than acute GVHD were higher center activity (HR = 1.8; 95% CI, 1.3–2.4; $P < 0.001$), and absence of *in-vivo* T-cell depletion with ATG (HR = 2.4; 95% CI, 1.7–3.3; $P < 0.001$) or with alemtuzumab (HR = 2.4; 95% CI, 1.6–3.7; $P < 0.001$). In multivariate time-dependent analyses, occurrence of limited chronic GVHD was associated with a trend for a lower risk of relapse (HR = 0.7; 95% CI, 0.5–1.0; $P = 0.07$), a suggestion for higher NRM (HR = 1.5; 95% CI, 0.9–2.3; $P = 0.4$), and significantly improved OS (HR = 1.8; 95% CI, 1.3–2.4; $P < 0.001$; Figure 2), while occurrence of extensive chronic GVHD was associated with a lower risk of relapse (HR = 0.7; 95% CI, 0.5–0.9; $P = 0.02$), but higher NRM (HR = 3.5; 95% CI, 2.4–5.2; $P < 0.001$), and comparable OS (HR = 1.1; 95% CI, 0.8–1.4; $P = 0.58$). There was no interaction between the disease status at transplantation nor the donor type and the impact of GVHD subtypes on transplantation outcomes. Interestingly, 3-year NRM (26 ± 3% versus 27 ± 6%; $P = 0.80$) and OS (61 ± 3% versus 63 ± 6%; $P = 0.78$) from diagnosis of extensive chronic GVHD were comparable in patients transplanted in centers with higher or lower activity.

To further assess the GVL effect of either chronic GVHD or of both acute and chronic GVHD, we performed two landmark analyses in patients who were leukemia-free at 18 months after transplantation ($n = 776$). In the landmark analysis looking at the impact of chronic GVHD, patients with chronic GVHD before the landmark day had a lower relapse rate ($P < 0.001$), higher NRM ($P < 0.001$) and similar survival than those without chronic GVHD before the landmark day (Figure 3). For the landmark analysis looking at the impact of both acute and chronic GVHD, patients were separated into three

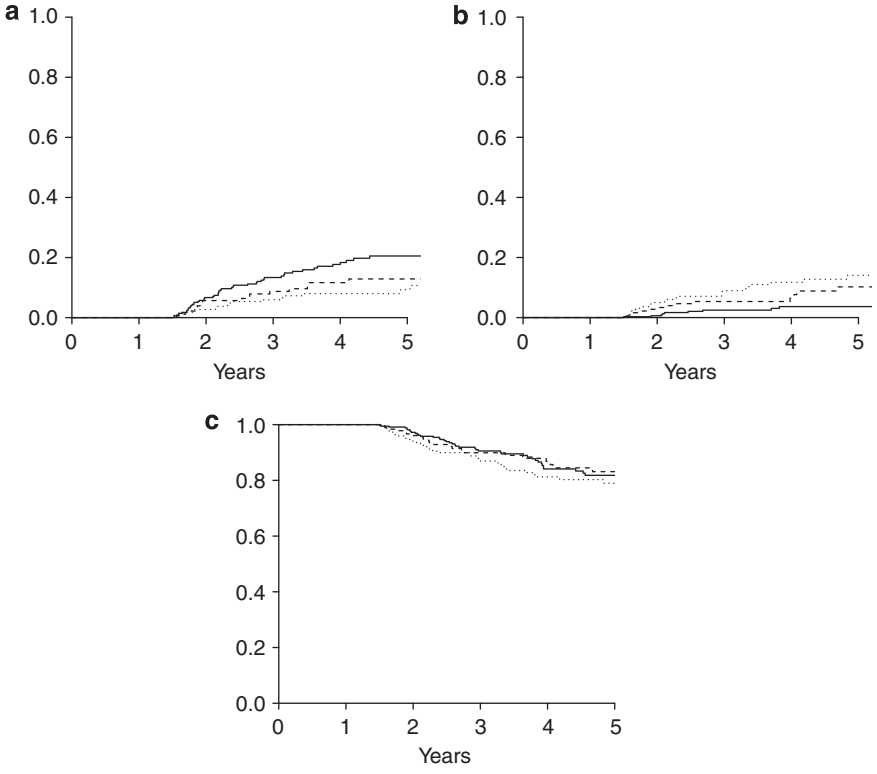

Figure 3. (**a**) Landmark analysis at 18 months after transplantation showing the cumulative incidence of relapse in patients who had no chronic GVHD (continuous line), limited chronic GVHD (broken line - - -) or extensive chronic GVHD (dotted line) before 18 months after RIC allo-SCT. Median follow-up from this landmark day was 24 (range, 0.1–112) months. (**b**) Landmark analysis at 18 months after transplantation showing the cumulative incidence of NRM in patients who had no chronic GVHD (continuous line), limited chronic GVHD (broken line - - -) or extensive chronic GVHD (dotted line) before 18 months after RIC allo-SCT. (**c**) Landmark analysis at 18 months after transplantation showing OS in patients who had no chronic GVHD (continuous line), limited chronic GVHD (broken line - - -) or extensive chronic GVHD (dotted line) before 18 months after RIC allo-SCT.

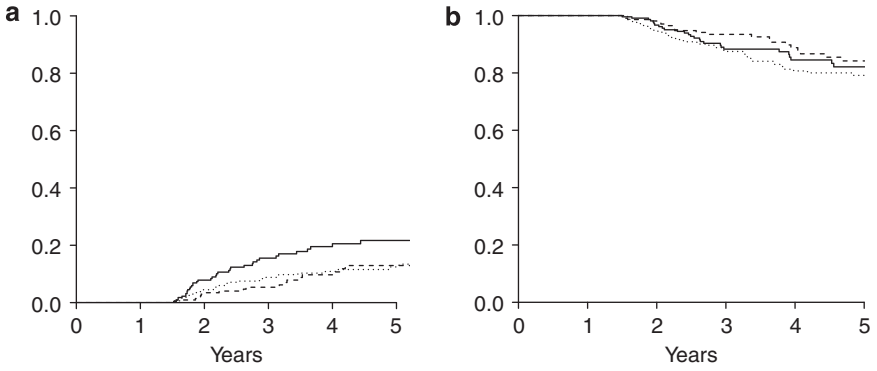

Figure 4. (**a**) Landmark analysis at 18 months after transplantation showing the cumulative incidence of relapse in patients who had no GVHD (continuous line) (at 4 years = 20 ± 3%), mild GVHD (broken line — —; defined as grade I acute or limited chronic GVHD) (at 4 years = 10 ± 3%) or severe GVHD (dotted line - - -, defined as grade II–IV acute and/or extensive chronic GVHD) (at 4 years = 10 ± 2%) before 18 months after RIC allo-SCT. (**b**) Landmark analysis at 18 months after transplantation showing OS in patients who had no GVHD (continuous line) (at 4 years = 85 ± 3%), mild GVHD (broken line — —) (at 4 years = 88 ± 3%) or severe GVHD (dotted line- - -) (at 4 years = 81 ± 2%) before 18 months after RIC allo-SCT.

groups: a group of patients without any GVHD (no GVHD group), a group of patients with grade II–IV acute GVHD and/or with extensive chronic GVHD (severe GVHD group), and a third group including remaining patients (mild GVHD group). As shown in the Figure 4, 4-year OS were 88 ± 3% in patients with mild chronic GVHD, 85 ± 3% in patients without GVHD and 81 ± 2% in patients with severe GVHD.

Impact of *in-vivo* depletion with ATG or alemtuzumab on transplantation outcomes

In multivariate analyses, *in-vivo* T-cell depletion with ATG was associated with a higher risk of relapse (HR = 1.5; 95% CI, 1.2–2.0; $P = 0.008$), a trend for lower NRM (HR = 0.7; 95% CI, 0.5–1.0; $P = 0.08$), similar LFS (HR = 0.8; 95% CI, 0.7–1.0; $P = 0.11$) and similar OS (HR = 0.9; 95% CI, 0.7–1.2; $P = 0.51$), while *in-vivo* T-cell depletion with alemtuzumab was associated with a higher risk of relapse (HR = 1.5; 95% CI, 1.1–2.1; $P = 0.008$), a trend to a higher risk of NRM (HR = 1.4; 95% CI, 1–2.2; $P = 0.08$), a lower LFS (HR = 0.7; 95% CI, 0.5–0.8; $P = 0.001$) and a lower OS (HR = 0.6; 95% CI, 0.5–0.8; $P = 0.001$) (Table 2). Finally, there was no interaction between the disease status at transplantation nor the donor type and the impact of *in-vivo* T-cell depletion on transplantation outcomes.

DISCUSSION

AML control after RIC allo-SCT has been in part attributed to the immune-mediated GVL effect.[28] Given the close relationship between GVHD and GVL, this study sought to assess the impact of GVHD on outcome in a large and rather homogeneous cohort of AML patients given PBSC after various RIC, and particularly at assessing the impact of GVHD on survival given the divergent results observed in studies containing fewer number of patients.[10–12,20–24] Several observations could be drawn from the current findings.

First, current study observed a positive impact (that is, lower risk of relapse translating into better LFS) of grade I acute GVHD on transplantation outcomes in patients given RIC allo-SCT for AML. This indirectly demonstrates the susceptibility of AML to graft-versus-tumor effects. In contrast, as observed by other groups of investigators,[10,20,21] current data indicated that grade II acute GVHD had no net impact on LFS/OS because the lower risk of relapse in patient with grade II–IV acute GVHD was offset by a higher NRM, and grade III–IV acute GVHD was associated with worse LFS/OS.

Second, current data confirmed an association between chronic GVHD and a lower risk of AML relapse, as observed previously by other groups of investigators,[10,11] but demonstrated that the beneficial effect of chronic GVHD in terms of survival was restricted to patients with limited chronic GVHD, given that extensive chronic GVHD was strongly associated with higher NRM. As relapse and NRM are competing events, it cannot be excluded that a part of the lower risk of relapse in patients with extensive chronic GVHD was due to the competition between the risks of relapse and NRM rather than to a true antileukemic activity of extensive chronic GVHD.[29]

Another important finding or our study was that attempts at preventing extensive chronic GVHD by *in-vivo* T-cell depletion with ATG or alemtuzumab, although successful at preventing it, failed to improve OS/LFS owing to a significant increase in the incidence of relapse, even after adjusting for acute and chronic GVHD, suggesting that donor T cell could mediate GVL effects in the absence of clinical GVHD, perhaps through graft-versus-host reactions limited to the recipient hematopoietic system. While ATG use did not affect OS, the use of alemtuzumab was associated with significantly worse survival, because it did not protect from NRM. Unfortunately, given the retrospective nature of the study, we did not have data related to alemtuzumab or ATG dosages for all patients who were given an alemtuzumab-based or an ATG-based conditioning. Current results are in accordance with a recent study from the CIBMTR analyzing data from 1676 patients given grafts after RIC as treatment for various hematological malignancies demonstrating that the use of alemtuzumab and/or ATG was associated with higher risk of relapse, that translated into worse disease-free survival.[30]

Patients transplanted in high-activity centers had higher incidence of chronic GVHD and better outcomes owing to both lower relapse risk and lower NRM than those transplanted in lower-activity centers, as previously observed by Giebel *et al.*[27] The higher incidence of chronic GVHD in patients transplanted in high-activity centers (even after adjusting for the use of ATG or alemtuzumab) could be due in part at a better recognition of signs of chronic GVHD in larger transplant centers, or to different modulation strategies of postgrafting immunosuppression according to disease risk and minimal residual disease data after transplantation in patients transplanted in high-activity centers (leading to both lower risk of relapse and higher incidence of chronic GVHD). Interestingly, NRM in patients diagnosed with extensive chronic GVHD was similar in patients transplanted in centers with high or low activity, stressing that treatment of extensive chronic GVHD has remained a difficult challenge even in large transplant centers.

This study also identified poor risk cytogenetics, being in second versus in first CR, and conditioning with low-dose TBI-based RIC as being associated with worse LFS or OS, confirming previous reports.[18,31,32] Finally, our study demonstrated higher mortality in patients above 56 years of age at transplantation in comparison with younger patients, in contrast to what has been observed in a recent report from the CIBMTR analyzing data from 545 patients with AML given RIC allo-SCT,[18] perhaps because analyses were not adjusted for comorbidities at transplantation (more likely to be more frequent in older patients) in current study.

In conclusion, in this large cohort of AML patients transplanted in CR using G-CSF-mobilized PBSCs, occurrence of grade I acute GVHD was associated with a lower incidence of relapse that translated toward better LFS, while occurrence of chronic GVHD was associated with a lower risk of relapse that translated toward improved OS in patients with limited chronic GVHD but not in those with extensive chronic GVHD who experienced higher long-term NRM. Overall, these findings highlight the close relationship between graft-versus-host reactions and the potential benefit of the immune-mediated GVL effect in the RIC allo-SCT setting, but also underline the need for improving the prevention and treatment of severe GVHD in patients receiving RIC allo-SCT, perhaps through promoting regulatory T cells.[33,34]

CONFLICT OF INTEREST

MM has received lectures honoraria and research support from Genzyme and Fresenius whose products are discussed in this manuscript. FB has received lectures honoraria and research support from Genzyme. DB has received honoraria from Genzyme. AN has received research grants from Fresenius and Genzyme, took part in some of their clinical trials (Fresenius and Genzyme), participated in their investigator meeting and advisory board (Genzyme) and lectured for them (satellite meeting and special meeting + honorarium).

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Supplementary Information accompanies the paper on the Leukemia website (http://www.nature.com/leu)