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Umbilical Cord Blood Cytomegalovirus Serostatus Does Not Have an Impact on Outcomes of Umbilical Cord Blood Transplantation for Acute Leukemia

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A B S T R A C T

Several studies have reported an impact of adult hematopoietic stem cell donor cytomegalovirus (CMV) serostatus on allogeneic hematopoietic cell transplantation outcomes. Limited data, however, are available on the impact of cord blood unit (CBU) CMV serostatus on allogeneic umbilical cord blood transplantation (UCBT) outcomes. We analyzed, retrospectively, the impact of CBU CMV serostatus on relapse incidence (RI) and 2-year nonrelapse mortality (NRM) of single-unit CBU transplantation for acute leukemia. Data from 1177 de novo acute leukemia pediatric and adult patients transplanted within European Group for Blood and Marrow Transplantation centers between 2000 and 2012 were analyzed. CBUs were provided by the European Cord Blood Banks. The median follow-up time for live patients was 59.9 months. The recipients of CMV-seropositive and -seronegative CBUs showed a comparable RI (33% versus 35%, respectively, $P = .6$) and 2-year cumulative incidence of NRM (31% versus 32%, respectively, $P = .5$). We conclude that CBU CMV serostatus did not influence RI and NRM in de novo acute leukemia patients after allo-UCBT and should not be included as a criteria for cord blood choice.

INTRODUCTION

Cytomegalovirus (CMV) status of the graft in the context of allogeneic hematopoietic stem cell transplantation (HSCT) has been described to impact on transplant outcomes. Both CMV seropositivity (ie, presence of anti-CMV IgG class antibodies) of the recipient and transplantation of seropositive recipient with a seronegative stem cell donor are known risk factors for increased transplant-related mortality with respect to allogeneic bone marrow or peripheral blood HSCT [1-5]. It may be related to the direct or indirect effect of the virus, CMV disease, toxicity of antiviral treatment, impaired engraftment, and worsening graft-versus-host disease (GVHD) [6-8]. However, several studies have demonstrated reduction

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in relapse risk after adult related and unrelated donor HSCT associated with CMV reactivation in both adult and pediatric patients with hematologic diseases [9,10]. The exact biologic mechanism(s) between CMV reactivation and decreased relapse is not yet fully understood. One hypothesis is a potential anti-leukemic effect of adaptive natural killer cells driven by the CMV reactivation post-HSCT [11]. Conversely, a very recent large retrospective study from the Center for International Blood and Marrow Transplant Research showed that CMV reactivation had no preventative effect on hematologic disease relapse [5].

In umbilical cord blood transplant (UCBT) recipients, the data on the cord blood unit (CBU) CMV serostatus is scarce. The incidence of a congenital CMV infection is low, approximately .5% to 2.0% of neonates, depending on socioeconomic and geographic factors [12]. Thus, CMV antibody positivity in cord blood generally reflects the mother's lifetime exposure due to the passive transport of maternal IgG antibody across the placenta. However, there is some evidence that during pregnancy the fetus may be exposed to the CMV antigens present in latent stage in the mother, even if the mother shows only IgG antibody positivity and with no symptoms of CMV infection (Paul Griffiths, personal communication). One study showed no association between CBU CMV serostatus and incidence of post-transplantation CMV infection [13]. Although the CBU T cells have always been considered naive (antigen inexperienced) compared with peripheral blood and bone marrow grafts [14,15], laboratory data published lately reports refuted this idea. It is possible that fetal and adult T cells are different. In fact, it was demonstrated that under stimulation, naive T cells can transform more quickly than adult T cells into functionally active central memory ($CD4^+CD45RO^+$) T cells in vitro [16].

Later, these data were complimented by clinical observations. Pediatric recipients of UCBT had higher naive $CD4^+$ T cells and TCR diversity compared with recipients of other grafts and early thymic-independent peripheral $CD4^+$ T cell expansion, with a rapid shift from naive to central memory phenotype observed in pediatric recipients of T cell–replete UCBT for malignant and nonmalignant disorders [17,18]. Furthermore, the ability to generate virus-specific T cells that target multiple viruses such as CMV, adenovirus, and Epstein-Barr virus from naive T cell populations in CBU confirms the ability of these cells to function [19]. Moreover, even more natural killer cell precursors were described in CBU with more rapid and sustained expansion [20,21]. Thus, fetus exposure during pregnancy to maternal CMV antigens present in the latent stage may lead to an early activation of the fetal immune system and generation of an immune response, with potential role in mediating the graft-versus-leukemia effect post-UCBT. With the aim to analyze the impact of CBU CMV serostatus on outcomes after UCBT, we conducted a retrospective analysis of 1177 pediatric and adult patients with acute leukemia reported to Eurocord.

METHODS

Subjects

Participants were children (ie, age ≤ 18 years) and adults with primary acute lymphoid leukemia or acute myeloid leukemia who underwent allogeneic UCBT after a myeloablative conditioning (MAC) or reduced-intensity conditioning (RIC) regimen in European Group for Blood and Marrow Transplantation centers between January 1, 2000 and December, 31 2012. Recipients who had undergone previous allogeneic HSCT or double-unit UCBT were excluded for the purpose of homogeneity. Patients who received a CBU provided from non–European Cord Blood Banks were also excluded. A total of 1177 patients were considered for this study.

Data Collection

Data on selected CBUs and their maternal characteristics were obtained from European Cord Blood Banks through a questionnaire on maternal and CBU characteristics. All participating Cord Blood Banks received the synopsis of the study, and 33 gave their consent (the list of participating Cord Blood Banks is in Acknowledgments). Data on patients, transplant procedures, and outcomes were cryopreserved from the electronic Eurocord database. All data were validated and checked by data managers and physicians in each European Group for Blood and Marrow Transplantation transplant center and Eurocord medical coordinator. The Institutional Review Board of Eurocord approved the study.

Statistical Analyses and Definitions

The primary endpoints were relapse incidence (RI), defined on the basis of morphologic evidence of acute leukemia in bone marrow, blood, or extramedullary organs, and nonrelapse mortality (NRM), defined as the death without relapse. The secondary endpoints were overall survival (OS), defined as the time between the date of transplantation and the date of death from any cause or last observation alive, and disease-free survival (DFS), defined as the time from transplantation to relapse, death, or last follow-up. Other endpoints were incidence of neutrophil engraftment at 60 days, defined as first of 3 consecutive days with a neutrophil count $\geq .5 \times 10^9$/L, and incidence of acute (aGVHD) and chronic (cGVHD) GVHD. aGVHD and cGVHD were defined and graded according to the previously published criteria [22,23]. HLA compatibility was determined at the antigen level for HLA-A and HLA-B loci and at the allele level for the HLA-DRB1 locus.

Cumulative incidence of neutrophil engraftment, NRM, and cGVHD were analyzed as the incidence rates and their 95% confidence intervals (CIs) to account for competing risks. Probabilities of OS and DFS were calculated using the Kaplan-Maier method, and the log-rank test was used for univariate comparisons. Multivariate analyses were performed using Cox proportional hazard regression model for DFS and OS and Fine and Gray's proportional hazard regression model for neutrophil engraftment, RI, NRM, and cGVHD. Because of the significant amount of missing data on date of onset of aGVHD, univariate analysis for factors associated with aGVHD was conducted using proportions, whereas logistic regression analysis was used for multivariate analysis.

Variables that reached $P = .2$ in the univariate analysis were included in the initial models. CBU CMV serostatus (determined by the maternal CMV) was included in all multivariate model cases regardless of the P value obtained in the univariate analysis. Variables were eliminated 1 by 1 in a backward stepwise fashion to ensure only those that reached a $P = .05$ in the final model were included. Other factors considered were recipient's age, gender, CMV status, diagnosis, and stage of disease at UCBT; HLA compatibility (6/6 and 5/6 versus 4/6 and 3/6); median number of cryopreserved total nucleated cells and median of cryopreserved $CD34^+$ cells; conditioning regimen (MAC versus RIC); and serotherapy (antithymocyte globulin [ATG]) use.

Statistical analyses were performed using IBM SPSS Statistics 20 (IBM Corp., Armonk, NY) (Copyright IBM Corporation 1989, 2011) and R 2.14.0 (Copyright R Foundation for Statistical Computing, Vienna, Austria, 2011) software packages. For all tests, 2-sided $P < .05$ was considered significant.

RESULTS

Patients and Transplant Characteristics

Included in this study were 1177 patients. Patients and transplant characteristics are summarized in Table 1. CMV seropositivity was reported in 56% of patients (n = 654) and 52% of CBUs (n = 613). Median follow-up time for survivors was 59.9 months (range, .1 to 183).

Relapse and NRM

Overall, 352 patients relapsed (of whom 312 died), with a median time to relapse of 8.2 months (range, 1 to 183). Maternal CMV status was not associated with relapse ($P = .7$), Figure 1A. The cumulative incidence of relapse at 5 years was 33.8% (95% CI, 31 to 37). RI was 30% (95% CI, 27 to 33) for patients in hematologic remission and 54.5% (95% CI, 47 to 63) for patients who presented with advanced-stage disease before UCBT ($P < .001$). In a multivariate analysis, risk of relapse was higher in patients with advanced disease at time of UCBT (hazard ratio [HR], 2.4; 95% CI, 2.1 to 2.8; $P < .001$), RIC regimen before transplant (HR, 1.3; 95% CI, 1.1 to 1.5; $P = .01$), and ATG use (HR, 1.4; 95% CI, 1.3 to 1.5; $P = .007$). Maternal CMV